

RELATION BETWEEN OXIDATIVE STRESS AND CAROTID INTIMA-MEDIA THICKNESS IN BETA-THALASSEMIA MAJOR PATIENTS

Thesis Submitted for fulfillment of Ph.D. Degree in Childhood Studies (Child Heath and Nutrition) Medical Studies Department

Presented by

Hany Shawky Mortagy

M.B.,B.Ch, M.Sc. in Pediatrics

Ain Shams University

Supervised by

Prof.Dr. Maisa Naser Farid **Prof.Dr. Randa Mahmoud Professor of Pediatrics** Asaad Sayed Matter Institute of Postgraduate Childhood Studies **Professor of Pediatrics** Ain Shams University Faculty of Medicine Ain Shams University **Prof.Dr. Manal Ahmed Shams-Eldien** Dr. Omneya Ibrahim Youssef **Professor of Clinical Pathology** Lecturer of Pediatrics Faculty of Medicine Faculty of Medicine Ain Shams University Ain Shams University 2014





Contents

	Page
Abstract	i
List of Abbreviations	iv
List of Tables	v
List of Figures	viii
Introduction	3
Aim of the Study	7
Review of Literature	11
Normal Human Hemoglobin	11
Thalassemia Syndrome	14
Oxidative Stress & Antioxidants	51
Glutathione Peroxidase	55
Nitric Oxide	59
Total Antioxidant Capacity (TAC)	66
Carotid Intima Media Thickness	74
Patients and Methods	85
Results	93
Discussion	121
Summary	129
Recommendations	133
References	137
Arabic Summary	١

List of Abbreviations

BMT	Bone Marrow Transplantation
CD	Clusters of differentiation
CRP	C-Reactive protien
СТ	Computed Tomography
EC	Endothelial cells
GH	Growth Hormone
DFO	Desferroxamine
GPX	Glutathione Peroxidase
Hb A	Hemoglobin A
Hb E	Hemoglobin E
Hb F	Hemoglobin F
Ig A	Immunoglobulin A
Ig G	Immunoglobulin G
Ig M	Immunoglobulin M
MR	Magnitic Resonance
MRI	Magnitic Resonance Imaging
NASH	Non Alcholic Steatohepatitis
NO	Nitric Oxide
РТ	Prothrombin time
PTT	Partial Thromboplastin Time
RBC	Red Blood Corpuscle
S.TFR	Serum Transferrin Receptors

Jist of Abbreviations

SCD	Sickle Cell Disease
TAC	Total Antioxidant Capacity
TI	Thalassemia Intermedia
β_ΤΜ	Beta Thalassemia Major



List of Tables

	Title	Page
Table (1):	Clinical and hematologic features of	
	the principal forms of thalassemias	17
	(Honig, 2004).	
Table (2):	Pharmacokinetic and clinical	
	characteristics of three iron chelators.	45
Table (3):	Main Reactive Oxygen Species (ROS)	
	in Superior Organisms.	52
Table (4):	Common Physiological Antioxidants	
	in Human Fluids	53
Table (5):	Total Antioxidant Capacity values in	
	foods (µmol Trolox-Equivalent/100g or	73
	100ml).	
Table (6):	Descriptive data of all thalassemic	
	patients	93
Table (7):	Descriptive data of control group	94
Table (8):	Comparison between thalassemic	
	patients and controls	95
Table (9):	Comparison between thalassemic	
	patients and control group as regards	96
	weight percentile	
Table (10):	Comparison between thalassemic	
	patients and controlgroup as regards	98
	height percentile	

List of Tables & Figures

	Title	Page
Table (11):	Comparison between thalassemic patients and control group as regards body mass index percentile	99
Table (12):	Correlation between CIMT and other parameters in thalassemic patients:	101
Table (13):	Correlation between TAC and other parameters in thalassemic patients	102
Table (14):	Correlation between Glutathione peroxidase and other parameters in thalassemic patients	103
Table (15):	Correlation between Nitric Oxide and other parameters:	105
Table (16):	Comparison between thalassemic patients on Desferoxamine and Deferiprone	106
Table (17):	Comparisonbetweenthalassemicpateints on Desferoxaminetherapy andDeferipronetherapy as regards sex	108
Table (18):	Comparison between thalassemic patients on Desferoxamine and on deferiprone therapy as regards weight percentile:	109
Table (19):	Comparison between Thalassemic patients on Desferoxamine therapy and Deferiprone therapy as regards height percentile:	111



	Title	Page
Table (20):	Comparison between thalassemic patients on Desferoxamine and on deferiprone therapy as regards body mass index:	113
Table (21):	Comparison between Thalassemic patients with serum ferritin below 2500 mg/dl and patients with serum ferritin above 2500 mg/dl:	115
Table (22):	Comparison between splenectomised and non splenectomised thalassemic patients.	116



List of Figures

	Title	Page
Fig. (1):	α , β globin genes on chromosome 11,16	11
	$[\alpha = alpha, \beta = beta, \gamma = gamma, \delta =$	
	delta, $\varepsilon = epsilon$, $\zeta = zeta$] (<i>Honig</i> , 2004)	
Fig. (2):	The composition of embryonic, fetal, and	13
	adult Hb (α = alpha, β – beta, γ = gamma,	
	δ = delta, ϵ = epsilon, ζ = zeta) (<i>Honig</i> ,	
	2004)	
Fig. (3):	Proportions of the various human Hb	13
	polypeptide chains through early life	
	(Honig, 2004).	
Fig. (4):	The geographical distribution of the	15
	thalassemias and the more common,	
	inherited structural hemoglobin	
	abnormalities (Hoffbrand et al., 2001	
Fig. (5):	Examples of mutations which produce β -	22
	thalassemia.FS=frame-shifts,	
	NS=nonsense, SPL=splicing (Hoffbrand	
	et al., 2001)	
Fig. (6a):	The facial appearance of a child with β -	33
	thalassemia major.	
Fig. (6b):	The skull x-ray in β -thalassemia major	33
	(Lilleyman et al., 2000).	

List of Tables & Figures

	Title	Page
Fig. (7):	A flowchart showing an approach to the	34
	diagnosis of the thalassemia syndromes	
	(Weatherall, 1995).	
Fig. (8):	Rickets like radiological lesions in	41
	thalassemia major patients caused by	
	desferoxamine (Neufeld, 2006)	
Fig. (9):	Pathophysiology and prevention of Total	71
	Antioxidant Capacity decay. (Arrows:	
	stimulation, Head arrow: inhibition)	
	(Kusano and Ferrari 2008)	
Fig. (10):	Schematic showing the anatomy and sites	76
	of intima-media arterial wall thickness	
	evaluation in the carotid system	
	(Mookadam et al., 2010)	
Fig. (11):	Descriptive data of control group	94
Fig. (12):	Comparison between thalassemic patients	96
	and controls	
Fig. (13):	Comparison between thalassemic patients	97
	and control group as regards weight	
	percentile	
Fig. (14):	Comparison between thalassemic patients	
σ	and control group as regards weight	
	nercentile	
	percentitie	



	Title	Page
Fig. (15):	Comparison between thalassemic patients	100
	and control group as regards body ass	
	index percentile	
Fig. (16):	Comparison between thalassemic patients on Desferoxamine and Deferiprone.	108
Fig. (17):	Comparison between thalassemic pateints	109
	on Desferoxamine therapy and	
	Deferiprone therapy as regards sex	
Fig. (18):	Comparison between thalassemic patients on	110
	Desferoxamine and on deferiprone therapy as	
	regards weight percentile.	
Fig. (19):	Comparison between Thalassemic	112
	patients on Desferoxamine therapy and	
	Deferiprone therapy as regards height	
	percentile.	
Fig. (20):	Comparison between thalassemic patients	114
	on Desferoxamine and on deferiprone	
	therapy as regards body mass index.	
Fig. (21):	Comparison between splenectomised and	117
	non splenectomised thalassemic patients	

<u>Acknowledgment</u>

I wish to express my deepest thanks, unlimited dept of gratitude, sincere respect, grateful acknowledgment to **Prof. Dr. Maisa Naser Farid** Professor of Pediatrics, Inistitute of Postgraduate Childhood Studies-Ain Shams University, for her continuous support, dead interest during all phases of the research, constant encouragement and unlimited help.

I would like to present my sincere thanks, appreciation and deepest gratitude to **Prof. Dr. Randa Matter,** Professor of Pediatrics, Faculty of Medicine, Ain Shams University, for her kind supervision, her great help through her vast experience and valuable suggestions.

I am deeply grateful for **Prof. Dr. Manal Ahamed Shams-Aldien Eltelbany,** Professor of Clinical Pathology, Faculty of Medicine, Ain Shams University, for her generous co-operation to achieve this work.

I would like to present my sincere thanks to $\mathcal{D}r$. *Omneya Ibrahim Yossef* lecturer of Peditric Faculty of Medicine, Ain Shams University for her great help to achieve her work.



ABSTRACT

Background: Beta-thalassemia is a heridetary hemolytic anemia with varying degrees of severity. Severely affected patients are treated with blood transfusion to maintain optimum level of hemoglobin for normal growth and physical activities. Vascular dysfunction with increase arterial stiffness and endothelial dysfunction has been demonstrated in patients with beta-thalassemia major patients.

Objective: The present study was carried out to explore the relation between oxidative stress markers in beta-thalassemia major patients and carotid intima-media thickness.

Methodology: The study included 40 thalassemic patients recruited from Hematology-Clinic Faculty of Medicine – Ain Shams University in addition to 50 healthy children who served as control group. All cases were subjected to full history taking, clinical examination and laboratory investigations including CBC, liver functions, serum ferritin, Hb electrophoresis, NO, TAC and glutathione peroxidase. CIMT were measured to both patrients and controls.

Results: There was a highly significant decrease in nitric oxide and total antioxidant capacity in thalassemic patients in comparison with control group. There was a highly significant increase in glutathione peroxidase and carotid intima-media thickness in thalassemic patients in comparison with control group. The study concluded a significant increase in carotid



intima-media thickness (CIMT) in splenectomised thalassemia major patients in comparison with non splenectomised patients.

Conclusion: There is a relation between oxidative stress markers in beta-thalasemia major patients and carotid intima-media thickness.

Keywords: CIMT, GPx, NO, TAC, Thalassemia major.



Aim of the Study

The present study was carried out to explore the relation between oxidative stress markers in β -thalassemia major patients and carotid intima-media thickness.