

# **Magnetic Resonance spectroscopy in patients with Thalassemia Major**

## ***Thesis***

*Submitted for Partial Fulfillment of Master Degree in  
Pediatrics*

## ***By***

***Soha Ahmed Hussien Ahmed***  
*M.B.,B.ch., (2007)*

## ***Supervisors***

**Dr. Rasha Hussien Aly Hussien**

*Asistent Professor of Pediatrics  
Faculty of Medicine, Ain Shams University*

**Dr. Khaled A. Ahmed**

*Asistent Professor of Diagnostic Radiology  
Faculty of Medicine, Ain Shams University*

**Dr. Rania Hamed Shatla**

*Asistent Professor of Pediatrics  
Faculty of Medicine, Ain Shams University*

*Faculty of Medicine, Ain Shams University*

2014

# Acknowledgment

*First and foremost, I feel always deeply indebted to **Allah**, the Most Gracious and the Most Merciful.*

*I would like to express my deepest gratitude and cardinal appreciation to **Prof. Rasha Hussien Ali Hussien**, Professor of Pediatrics, who kindly supervised and motivated the performance of this work, for her kind guidance and constant encouragement throughout this work.*

*I am greatly honored to express my sincere appreciation to **Prof. Khaled A. Ahmed**, Professor of Diagnostic Radiology, for his continuous support, help and generous advice throughout this work.*

*I am deeply thankful to **Dr. Rania Hamed Shatla**, Lecturer of Pediatrics for her great help, outstanding support, active participation and guidance.*

*Finally, I want to dedicate this work to all the members of my family because of their patience and support.*

*Soha Ahmed Hussien Ahmed*

# *List of Contents*

Title	Page No.
List of Tables .....	i
List of Figures .....	iii
List of Abbreviations .....	v
Introduction .....	1
Aim of the Work.....	4
Review of Literature	
○ Thalassemia.....	5
○ Magnetic Resonance Spectroscopy (MRS) .....	42
Subjects and Methods .....	64
Results .....	69
Discussion .....	84
Summary .....	92
Conclusion.....	97
Recommendation .....	98
References .....	99
Arabic Summary	

## *List of Tables*

Table No.	Title	Page No.
<b>Table (1):</b>	Shows the level of brain metabolites in different types of tumors .....	49
<b>Table (2):</b>	Clinical data: as regards age, weight, height, BMI and sex. ....	69
<b>Table (3):</b>	Laboratory data among all studied cases .....	70
<b>Table (4):</b>	Radiological findings .....	70
<b>Table (5):</b>	Clinical data; as regards age, age of diagnosis of Thalassemia,Error! Bookmark not defined.	
<b>Table (6):</b>	Iron chelating therapy:Error! Bookmark not defined.	
<b>Table (7):</b>	Laboratory data; as regards hemoglobin, serum ferritin level and alanine transaminase. .... Error! Bookmark not defined.	
<b>Table (8):</b>	Clinical data, as regards age, age of diagnosis, age of 1st blood transfusion, No. of blood transfusion, sex, family history, splenectomy, complications. ....	71
<b>Table (9):</b>	Iron chelating therapy: .....	72
<b>Table (10):</b>	Compliance to iron chelating therapy .....	72
<b>Table (11):</b>	Laboratory data, as regards hemoglobin, serum ferritin level and alanine transaminase. .... Error! Bookmark not defined.	
<b>Table (12):</b>	Wechsler Intelligence Scale for Children forth edition (WISC IV) .....	73
<b>Table (13):</b>	Benton visual Retention test.....	77
<b>Table (14):</b>	Winconsin card sort test.....	79

## *List of Tables (cont...)*

Table No.	Title	Page No.
<b>Table (15):</b>	Correlation between NAA/Cr Ratio and clinical data, as regards (age, age of diagnosis, age of 1st blood transfusion, No. of blood transfusion, sex, family history, splenectomy & complications).....	81
<b>Table (16):</b>	Correlation between NAA/Cr Ratio and Iron chelating therapy, as regards (type and compliance to iron chelating therapy).....	81
<b>Table (17):</b>	Correlation between NAA/Cr Ratio and Laboratory data, as regards (hemoglobin, serum ferritin level and ALT).Error! Bookmark not defined.	
<b>Table (18):</b>	Wechsler Intelligence Scale for Children forth edition (WISCIV).....	82
<b>Table (19):</b>	Benton Visual Retention Test (BVRT).....	82
<b>Table (20):</b>	Winconsin Card Sort Test (WCST) .....	83

## *List of Figures*

Fig. No.	Title	Page No.
<b>Figure (1):</b>	Hemoglobin structure.....	7
<b>Figure (2):</b>	Mechanism of ineffective erythropoiesis and hemolysis in thalassemia .....	9
<b>Figure (3):</b>	A thalassaemic patient showing characteristic facial appearance .....	13
<b>Figure (4):</b>	Skull X-ray of a child with b thalassemia major showing a 'hair on- end' appearance as a consequence of marked erythroid hyperplasia.....	26
<b>Figure (5):</b>	Management of thalassemia and treatment of related complications .....	41
<b>Figure (6):</b>	N-Acetylaspartate Structure .....	43
<b>Figure (7):</b>	Normal spectra in newborn .....	52
<b>Figure (8):</b>	Changes in metabolite concentrations with age .....	52
<b>Figure (9):</b>	Computerized version of Wechsler Intelligence Scale for Children.....	55
<b>Figure (10):</b>	A sample design used in Administration M of the Benton Test the four design choices are shown and the subject is asked to choose the one that best matches the original design .....	56
<b>Figure (11):</b>	Computerized version of the Wisconsin Card sort.....	58
<b>Figure (12):</b>	Shows MRS of one of our B-TM male patients, 10 years old presented with reduction of NAA/Cr ratio. ....	63
<b>Figure (13):</b>	Comparison between normal& reduced NAA/Cr ratio regards different levels of verbal I.Q. ....	75
<b>Figure (14):</b>	Comparison between normal& reduced NAA/Cr ratio regards different levels of performance I.Q.....	76

## *List of Figures (cont...)*

Fig. No.	Title	Page No.
<b>Figure (15):</b>	Comparison between normal & reduced NAA/Cr ratio regards different levels of total I.Q. ....	77
<b>Figure (16):</b>	Comparison between normal& reduced NAA/Cr ratio regards BVRT.....	78
<b>Figure (17):</b>	Comparison between normal & reduced NAA/Cr ratio regards WSCT.....	80
<b>Figure (18):</b>	Correlation between NAA/Cr ratio and Verbal I.Q showed a positive statistically relation.Error! Bookmark not defined.	
<b>Figure (19):</b>	Correlation between NAA/Cr ratio and performance I.Q showed a positive statistically relation.Error! Bookmark not defined.	
<b>Figure (20):</b>	Correlation between NAA/Cr ratio and total I.Q showed a positive statistically relation. .... Error! Bookmark not defined.	
<b>Figure (21):</b>	Correlation between NAA/Cr ratio and BVRT showed a negative statistically relation. .... Error! Bookmark not defined.	
<b>Figure (22):</b>	Correlation between NAA/Cr ratio and WSCT showed a positive statistically relation. .... Error! Bookmark not defined.	

## *List of Abbreviations*

Abb.	Meaning
Ala.....	Alanine
ALS .....	amyotrophic lateral sclerosis
BMI .....	Body mass index
BVRT .....	Benton Visual Retention Test
Cho.....	Choline
CNS .....	Central nervous system
Cr .....	Creatine
CVA .....	Cerebrovascular accident
DFO .....	Desferoxamine
DFP.....	Deferiprone
DFX .....	Deferasirox
DWMI .....	Deep white matter ischemia
Fe .....	Iron
GABA.....	Gamma-aminobutyric acid
Gln .....	Glutamine
Glu .....	Glutamate
Glx.....	Glutamate-Glutamine
GMDs.....	Glucose metabolic disorders
GVHD .....	Graft versus host disease
Hb .....	Hemoglobin
Hb F .....	Fetal hemoglobin
HbA1.....	Adult hemoglobin1
HbA2 .....	Adult hemoglobin2
HLA .....	Human leukocyte antigen
H-MRS .....	Proton MRS
HSCT .....	Hematopoietic stem cell transplantation
IQ .....	Intelligence quotient
Lac .....	Lactate
LIC.....	Liver iron content
Lip.....	Lipids
MRI .....	Magnetic resonance imaging
MRS .....	Magnetic resonance spectroscopy
MS .....	Multiple sclerosis



## *List of Abbreviations*

Abb.	Meaning
Myo .....	Myoinositol
NAA .....	N-acetylaspartate
NAA/Cho.....	N-acetylaspartate to Choline ratio
NAA/Cr .....	N-acetylaspartate to Creatine ratio
NAAG .....	<i>N</i> acetyl aspartyl glutamate
NMR .....	Nuclear Magnetic Resonance
PIH.....	Pyridoxal isonicotinoyl hydraxone
ppm .....	Parts per million
RBCs .....	Red blood cells
RDW .....	Red cell distribution width
SCD.....	Sickle cell disease
TE .....	Time to echo
UCB .....	Umbilical cord blood
VTEs .....	Venous thromboembolic events
WCST.....	Wisconsin Card Sorting Test
WISCIV.....	Wechsler Intelligence Scale for Children Forth edition
B-TI.....	Beta thalassemia intermediata
B-TM.....	Beta thalassemia major

## INTRODUCTION

**B**eta—Thalassemia is prevalent in Mediterranean countries. The Middle East, Central Asia, India, Southern China, and the Far East as well as countries along the total annual incidence of symptomatic individuals is estimated at 1 in 100,000 throughout the world and 1 in 10,000 people in European Union (*Galanello & Origa ., 2010*).

In Egypt, beta thalassemia is the most common type with a carrier rate varying from 5.3 % - 9 % and a gene frequency of 0.03. So, it was estimated that 1,000 1 /1.5 million per year live births will suffer from thalassemia disease in Egypt (total life births 1,936,205 in 2006 (*El-Beshilawy & Youssry ., 2009*)).

The life expectancy of children with B- TM is increased with transfusion, chelating therapy and bone marrow transplantation. In these patients, complications are generally associated with iron deposition resulting from frequent blood transfusions, hemolysis, and increased intestinal iron absorption, and thus pose a risk for parenchymal organ injuries (*Duman et al., 2010*).

Iron deposition mainly affects the liver, heart, pancreas, gonads, parathyroid and thyroid glands, bones, lungs, peripheral and central nervous system (CNS). CNS complications generally present as cognitive dysfunction, which usually results from iron deposition and neurotoxicity of deferoxamine (DFO), which is

commonly used as a chelating agent. Furthermore, hypoxia and thromboemboli may casuse CNS complications and cognitive dysfunction (*Duman et al., 2010*).

The side effects due to the disease itself or its treatment, being unable to attend school, frequent hospitalizations, and the physical & social restrictions as a consequence of chronic disease and its treatment also lead to cognitive dysfunction (*Duman et al., 2010*).

Children with untreated Thalassmia major have been reported to experience transient ischemic attacks, silent infarctions that result in brain injury, but often with subtle or undetectable clinical symptoms, and in rare cases not involving chronic transfusion therapy, stroke. A second risk factor, only recently recognized as significant for children with sickle cell disease, is the long-term effect of chronic anemia and associated hypoxia. The third risk factor relates directly to consequences of the treatment that has changed the natural course of this disease (*Armstrong et al., 2005*).

In thalassemic patients thrombo-embolic events has been described.

One of the affected organs is the brain where symptomatic and asymptomatic damage has been reported. Recent studies describe cases who preonted with the signs of cerebrovascular accident (CVA), some of them are isehemic

and others with hemorrhage or stroke. They not received regular blood transfusions (*Khanlarli et al., 1985*).

Magnetic resonance spectroscopy (MRS) offers a unique, noninvasive approach to assess pediatric neurological abnormalities at microscopic levels by quantifying cellular metabolites. The most widely available MRS method, proton spectroscopy, is approved for general use and can be ordered by clinicians for pediatric neuron imaging studies if indicated (*Panigraphy et al., 2010*).

MRS can detect changes in intracellular cerebral metabolites. For example MRS can detect decrease in N-acetyl-aspartate containing compounds in chronic hypoxia. Also decrease of choline level as hypoxia decreased choline phosphorylation, choline kinase activity so decrease choline containing compounds Also MRS can detect increase in lactate, glutamate & alanine levels when there is insufficient oxygen at the cellular level (*Bonavita & Disalle., 1999*).

Detection of NAA containing compounds by MRS in asymptomatic brain damage is indicated specially in group A patients with highly risk to develop thromboembolic events (*Khanlari et al., 1985*). As NAA is a marker of neuro/axonal integrity. NAA concentration in white matter is related to the structural and functional integrity of axonal fibers (*Aydin et al., 2012*).

## **AIM OF THE WORK**

**T**he aim of this study is uncovering the relation between neurocognitive impairment, serum ferritin and different iron chelators with cerebral intracellular metabolites by doing MRS to B-TM patients.

## Chapter (1)

# THALASSEMIA

**T**halassemia is an inherited autosomal recessive blood disease that originated in the Mediterranean region. In thalassemia the genetic defect, which could be either mutation or deletion, results in reduced rate of synthesis of one of the globin chains causing the formation of abnormal hemoglobin molecule (*Gulbis et al., 2009*).

### *Prevalence and Geographical Distribution:*

It has been estimated that about 1.5% of the global population (80 to 90 million people) are carriers of beta-thalassemia, with about 60,000 symptomatic individuals born annually, the great majority in the developing world (*Galanello and Origa, 2010*).

In Egypt, it is particularly common in populations of indigenous ethnic minorities of Upper Egypt and also peoples of the Nile Delta, Red Sea Hill Region and especially amongst the Siwan (*El-Beshlawy et al., 2007*).

### *Classification of $\beta$ -thalassemia:*

Beta-thalassemia is a genetically inherited hemoglobin disorder caused by impaired synthesis of the  $\beta$ -globin chain, which results in chronic hemolytic anemia. Currently, intensive blood transfusions and iron chelation therapy have improved

the life expectancy and reduced the incidence and severity of cardiac dysfunction and heart failure considerably (*Mokhtar et al., 2005*).

- 1-  **$\beta$ -thalassemia minor:** Heterozygous  $\beta$ - thalassemia is associated with no clinical abnormalities and may be mistaken for iron deficiency anemia(*Honig, 2004*).
- 2-  **$\beta$ -thalassemia intermedia ( $\beta$ TI):** About 10% of heterozygous  $\beta$ -thalassemia have a syndrome of intermediate hemolytic severity. Those patients usually have onset of anemia after 2 years of age and they do not require regular blood transfusion (*Yaish, 2007*).
- 3-  **$\beta$ -thalassemia major ( $\beta$ TM):** Homozygous  $\beta$ -thalassemia in which there is defective formation of  $\beta$  chain (*Honig, 2004*).

*There are 2 types:*

- a-  **$\beta^0$  thalassemia:** Complete absence of  $\beta$  chain.
  - b-  **$\beta^+$  thalassemia:**  $\beta$  chain synthesis is reduced.
- 4- **Other  $\beta$ -thalassemia syndromes:** Sick cell  $\beta$ -thalassemia (Hb-S- $\beta$ -thalassemia), hemoglobin C-  $\beta$ -thalassemia (Hb-C- $\beta$ -thalassemia) and hemoglobin D-  $\beta$  –thalassemia (Hb-D- $\beta$ -thalassemia).

(*Yaish, 2007*)