ASSESSMENT OF THE ROLE OF APOPTOSIS GENES IN MULTIPLE SCLEROSIS

Thesis

Submitted in Partial Fulfillment for
M.D. Degree in
NEUROLOGY

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FACULTY OF MEDICINE
CAIRO UNIVERSITY
2012

بِسْمِ اللّهِ الرَّحْمَنِ الرَّحِيمِ

"سُبْحَانَكَ لاَ عِلْمَ لَنَا إِلاَّ مَا عَلَّمْتَنَا
إِنَّكَ أَنتَ الْعَلِيمُ الْحَكِيمُ"

سورة البقرة(الآية ٣٢)

ACKNOWLEDGMENT

*First of all, I am deeply thankful to **Allah** by the grace of whom this work was possible.*

*This work was honored to be under supervision of Prof. **Mohammed Hussein El Tawdy**, Professor of Neurology, Faculty of Medicine, Cairo University, who gave his sincere supervision and support to the work all through.*

*It is my pleasure to express my deepest gratitude and sincere thanks to Prof. **Maged Mohammad Abdel Naseer**, Professor of Neurology, Faculty of Medicine, Cairo University, for his generous concern, sincere supervision, valuable suggestions and cooperation, continuous advice and support saving no effort or time in reading each word in this work.*

*I am also highly indebted to Prof. **Sanaa Sayed Abdel Shafy**, Professor of Clinical Pathology and Immunology, Faculty of Medicine, Beni Suef University, for her kind support, sincere supervision, advices and guidance throughout this work.*

*I wish also to express my sincere gratitude and thanks to Prof. **Mona Abdel Fatah Nada**, Assistant Professor of Neurology, Faculty of Medicine, Cairo University, for her kind supervision, sincere encouragement, continuous participation, valuable advices and instructions throughout this work.*

My deepest gratitude, appreciation and thanks to the members of the molecular biology and immunology lab of the critical care Department, Faculty of Medicine, Cairo University, for their help regarding the laboratory work in this study.

I would like to express my gratitude to all the members of neurology department for their help and support.

I am especially grateful to my family especially my mother and my husband for their endless patience and support throughout this work.

Amany Abolmagd

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LIST OF ABBREVIATIONS

2CdA	2-chlorodeoxyadenosine
ADA	Adenosine Deaminase
ALLEGRO	Assessment of Oral Laquinimod in Preventing Progression In Multiple Sclerosis
APAF-1	Apoptosis Protease-Activating Factor 1
APC	Antigen Presenting Cell
APO-3	Apolipoprotein-3
ATP	Adenosine Triphosphate
BAX	Bcl2 Associated X-protein
BBB	Blood–Brain Barrier
Bcl2	B-Cell Lymphoma 2
Bcl2-L	Bcl2-Like
Bcl-X_L	B-Cell Lymphoma-Extra Large
BCR	Bicaudate Ratio
BH3	Bcl2 Homology Domain 3
BID	Bcl2 Interacting Protein
BID	BH3 Interacting Death Domain
BIM	Bcl2-Interacting Protein
BRAVO	Benefit Risk Assessment of Avonex And Laquinimod
CAD	Caspase-Activated DNase
CCSVI	Chronic Cerebrospinal Venous Insufficiency
CD	Cluster Of Differentiation
CDC	Centers For Disease Control and Prevention

CDMS	Clinically Definite Multiple Sclerosis
CHMP	Committee For Medicinal Products For Human Use
CIAPs	Cellular Inhibitor of Apoptosis
CIS	Clinically Isolated Syndromes
CLARITY	Cladribine Tablets Treating Multiple Sclerosis Orally
CLEC16A	C-Type Lectin Domain Family 16
CMV	Cytomegalovirus
CNS	Central Nervous System
COX-2	Cyclooxygenase-2
CSF	Cerebrospinal Fluid
cyto-C	Cytochrome-C
DD	Death Domain
DED	Death Effector Domain
DHODH	Dihydroorotate Dehydrogenase
DISC	Death Inducing Signaling Complex
DMF	Dimethylfumarate
DNA	Deoxy Ribonucleic Acid
DRs	Death Receptors
EAE	Experimental Autoimmune Encephalomyelitis
EBNA1	EBV-Encoded Nuclear Antigen-1
EBV	Epstein–Barr Virus
EC	Extracellular
EDSS	Expanded Disability Status Scale
EDTA	Ethylenediaminetetraacetic Acid

ELISA	Enzyme-Linked Immunosorbent Assay
EMA	European Medicines Agency
Endo G	Endonuclease-G
FADD	Fas-Associated Via Death Domain
FAE	Fumarate
FAS	Factor For Apoptotic Signal
FasL	Fas Ligand
FDA	Food And Drug Administration
FLAIR	Fluid Attenuation Inversion Recovery
FLICE	FADD-Like IL-1 Beta-Converting Enzyme
FLIP	FLICE Inhibitory Protein
Foxp3	Forkhead Box Protein 3
FREEDOMS	FTY720 Research Evaluating Effects of Daily Oral Therapy in Multiple Sclerosis
FS	Functional System
FSS	Functional Systems Scores
FTY720	Fingolimod
GAPDH	Glyceraldehyde-3-Phosphate Dehydrogenase
GATA-3	GATA-Binding Protein 3
Gd	Gadolinium
HHV	Human Herpesvirus
HIV	Human Immunodeficiency Virus
HLA	Human Leucocyte Antigen
HTRA2	High Temperature Requirement Protein-A2
IAPs	Inhibitor Of Apoptosis Proteins

IC	Intracellular
ICAD	Inhibitor Of Caspase-Activated DNase
ICAM	Intracellular Adhesion Molecule
IFN-γ	Interferon Gamma
IG	Immunoglobulin
IL	Interleukin
IL-2R	IL-2 Receptor
IM	Infectious Mononucleosis
iNOS	Inducible Nitric Oxide Synthase
IRF8	Interferon Regulatory Factor 8
LAQ	Laqinimod
MAESTRO-01	A pivotal phase II, III trial for SPMS
MAESTRO-03	A pivotal phase III trial for SPMS for patients in the United States
MBP	Myelin Basic Protein
MDM2	Mouse Double Minute-2 Homolog
MHC	Major Histocompatibility Complex
MMF	Monomethyl Fumarate
MMP	Mitochondrial Membrane Permeabilization
MMPs	Metalloproteinases
MMR	Measles, Mumps and Rubella
MOG	Myelin Oligodendrocyte Glycoprotein
MRI	Magnetic Resonance Imaging
mRNA	Messenger Ribonucleic Acid
MRTC	Myelin Reactive T-Cells

MS	Multiple Sclerosis
MusiQoL	Multiple Sclerosis International Quality Of Life
NFκB	Nuclear Factor Kappa-Light-Chain-Enhancer of Activated B Cells
NK	Natural Killer
NKT-cells	Natural Killer T-Cells
Opg	Osteoprotegerin
OpgL	Osteoprotegrin Ligand
PBMC	Peripheral Blood Mononuclear Cells
PCR	Polymerase Chain Reaction
Pg/ml	Picogram Per Milliliter
PI	Progression Index
PLP	Proteolipid Protein
PPMS	Primary Progressive Multiple Sclerosis
PR	Primary Progressive
PTGER4	Prostaglandin E Receptor 4
PTK	Protein Tyrosine-Kinase
QoL	Quality Of Life
RANKL	Receptor Activator of NF-Kappa B Ligand
RGS1	Regulator Of G-protein Signaling 1
RNA	Ribonucleic Acid
RORγT	Retinoic Acid receptor Related Orphan Receptor
ROS	Reactive Oxygen Species
rpm	Revolutions Per Minute
RR	Relapsing-Remitting

RRMS	Relapsing-Remitting Multiple Sclerosis
RTL	Recombinant T Cell Receptor Ligand
RT-PCR	Real Time Polymerase Chain Reaction
S FAS	Soluble Fas
S1P	Sphingosine 1- Phosphate
SD	Standard Deviation
SMAC	Second Mitochondria-Derived Activator Of Caspase
SP	Secondary Progressive
SPMS	Secondary Progressive Multiple Sclerosis
STAT	Signal Transducer And Activator Of Transcription
T-bet	T-Box Expressed In T cells
tBID	Truncated BID
TCR	T Cell Receptor
TCV	T-Cell Vaccination
TF	Teriflunomide
TGF-β	Transforming Growth Factor -β
Th	T Helper
TNF	Tumor Necrosis Factor
TNFR	Tumor Necrosis Factor Receptor
TNFRSF1A	Tumor Necrosis Factor Receptor Superfamily Member 1A
TNFSF	Tumor Necrosis Factor Superfamily
TRADD	Tumor Necrosis Factor Receptor-1-Associated Death Domain
TRAIL	TNF-Related Apoptosis-Inducing Ligand

TRANSFORMS	Trial Assessing Injectable Interferon Versus FTY720 Oral in Relapsing–Remitting Multiple Sclerosis
Treg	T Regulatory Cells
TTV	Torque Teno Virus
UNG	Uracil DNA Glycosylase
USA	United States Of America
VCAM-1	Vascular Cell Adhesion Molecule- 1
VDR	Vitamin D Receptors
VEP	Visual Evoked Potential
VLA-4	Very Late Activation Antigen-4
VZV	Varicella-Zoster Virus
WHO	World Health Organization

ABSTRACT

Background: One of Multiple sclerosis (MS) presumed pathological mechanisms is failure of apoptosis of autoreactive T lymphocytes. **Objectives**: To determine the relationship between tumor necrosis factor-related apoptosis-inducing ligand (TRAIL) mRNA gene expression ratio and serum TRAIL levels with MS susceptibility and brain atrophy. **Materials and methods:** This study was conducted on 53 relapsing remitting MS patients and 25 matched healthy volunteers. The expression of TRAIL on peripheral blood lymphocytes was analyzed by RT-PCR, serum levels of soluble TRAIL (sTRAIL) was determined by ELISA and MRI brain for measurement of black holes and the bicaudate ratio (BCR) as a measure of brain atrophy were done for all patients. **Results:** The sTRAIL level was lower in MS patients compared to the control but no difference was found in the TRAIL mRNA gene expression ratio. A positive correlation was found between EDSS and age of patients while a negative correlation was found between progression index and both disease duration and BCR. **Conclusion:** Apoptosis of T lymphocytes is deficient in MS patients which can be implicated in the treatment. No difference between TRAIL mRNA gene expression ratio between MS patients and controls.

Keywords:

Multiple sclerosis, Apoptosis, Tumor necrosis factor related apoptosis inducing ligand (TRAIL), TRAIL mRNA gene expression, Bicaudate ratio.

INTRODUCTION

Multiple sclerosis (MS) is an immune mediated disease of the central nervous system (CNS) initiated by recruitment of activated T cells and macrophages to the brain **(McFarland & Martin, 2007).**

A failure of autoreactive immune cells to undergo apoptosis in MS may lead to inappropriate persistence of these cells and causes harmful immunoreactivity within the CNS **(Sharief, 2000; Saresella et al., 2005; Okuda et al., 2006; Wosik et al., 2007).**

Since the presence of apoptosis related molecules in the blood is thought to reflect the pathological process in the CNS, determination of these molecules may be useful in evaluation of disease activity **(Rinta et al., 2008).**

Tumor necrosis factor (TNF)-related apoptosis-inducing ligand (TRAIL) molecule is among the potential biomarkers in MS **(Wandinger et al., 2003; Gilli et al., 2006).**

TRAIL is a member of the tumor necrosis factor (TNF) superfamily with an immunomodulatory capacity having a protective role against autoimmunity. It inhibits T cell activation, cell cycle progression, interferon (IFN)-γ and interleukin (IL)-4 production. Since it can downregulate the T cell driven immune responses, TRAIL may carry a potential importance for modulating the outcomes of diseases **(Lünemann et al., 2002).**

A failure in apoptosis of autoreactive T cells may lead to increased immunoreactivity within the CNS that may damage myelin, oligodendrocytes, and neurons **(Zipp, 2000).**