The Reliability of Candida Skin Test in Investigating Tcell function in Infants

Thesis

Submitted for Partial Fulfillment of Master Degree

in Pediatrics

By

Shaimaa Abdel-Azim Mohamed El-Toukhy

Ain Shams University

Supervised by

Prof/ Shereen Medhat Reda

Professor of Pediatrics

Faculty of Medicine - Ain Shams University

Dr/ Rasha Hassan El-Owaidy

Lecturer of Pediatrics

Faculty of Medicine - Ain Shams University

Dr/ Neama Lotfy Mohamed

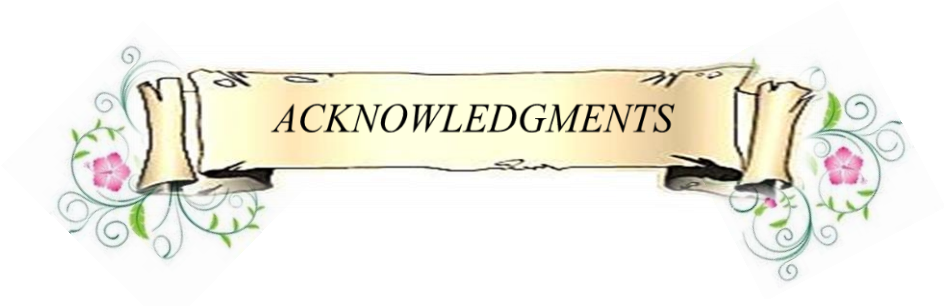
Lecturer of Clinical Pathology

Faculty of Medicine- Ain Shams University

Faculty of Medicine

Ain Shams University

2015



*First of all thanks to **ALLAH** the most merciful.*

*Secondly this dissertation would not have been possible without the help, support and patience of my principal supervisor of this thesis: **Prof. Shereen Medhat Reda**, Professor of Pediatrics at Faculty of Medicine - Ain Shams University,I would like to sincerely thank her for her outstanding support , supervision and expert guidance.*

*I would also like to sincerely thank **Dr. Rasha Hassan El-Owaidy**, Lecturer of Pediatrics, Faculty of Medicine - Ain Shams University, for her help, constructive criticism, elegant viewpoints and keen insights.*

*It is my pleasure also to thank **Dr. Neama Lotfy Mohamed**, Lecturer of Clinical Pathology, Faculty of Medicine - Ain Shams University for her time and for the great effort she has done for us.*

I really appreciate and would like to thank all the parents of our enrolled subjects for their cooperation and their trust in us

My thesis is dedicated to my parents, my husband, my son, my sister and brother; I wish to make them all proud of me.

Shaimaa Eltoukhy

Table of Contents

List of Abbreviations

ADA	:	adenosine deaminase assay
ALC	:	absolute lymphocyte count
ANC	:	absolute neutrophil count
APCs	:	antigen presenting cells
BCG	:	Bacillus Calmette-Guerin
C.albicans	:	candida albicans
CBC	:	complete blood count
CCR	:	chemokine receptor
CDs	:	cluster of differentiation
CFSE	:	carboxyfluorescin succinimidyl ester
CMF-PBS	:	calcium and magnesium free phosphate buffered saline
CO2	:	carbon dioxide
CON A	:	concanavalin A
Cr51	:	chromium 51 release assay
CTLs	:	cytotoxic T lymphocytes
CXCR5	:	cysteine-x-cysteine motif receptors 5
DN	:	double negative
DP	:	double positive
DTH	:	delayed type hypersensitivity
EDTA	:	ethylene diamine tetraacetic acid
ELISA	:	enzyme linked immunosorbent assay
ELISPOT	:	enzyme linked immunospot
FBS	:	fetal bovine serum
H1N1	:	hemagglutinin 1 and neuraminidase 1
Hb	:	Haemoglobin

HSC	:	haematopoietic stem cells
IL	:	Interleukin
INF- γ	:	interferon gamma
INF- γ R	:	interferon gamma receptor
IOR	:	interquartile ranges
JAK	:	janus kinases
LFα	:	lymphotoxin alpha
MCV	:	mean corpuscular volume
MHC	:	major histocompatibility complex
MNCs	:	mononuclear cells
MPPs	:	multipotent progenitors
mTECs	:	medullary thymic epithelial cells
NK	:	natural killer cells
OD	:	optical density
PBS	:	phosphate buffered saline
PHA	:	phytohemagglutinin
PLT	:	Platlets
PNL	:	polymorphonuclear leukocytes
PNP	:	purine nucleoside phosphorylase
PPD	:	purified protein derivative
RPMI	:	Roswell Park Memorial Institute
SCID	:	severe combined immunodeficiency
SD	:	standard deviation
STAT	:	signal transducer and activator of transcription
T reg	:	T regulatory cells
TCR	:	T cell receptor
TEM	:	effector memory T cell
Tfh	:	T follicular helper cells

TGF-beta	:	transforming growth factor beta
Tr1	:	type 1 regulatory T cell
TRECs	:	T cell receptor excision circles
TSAs	:	tissue specific self antigens
V(D)J	:	variable diversity joining
WBCs	:	white blood cells
ZAP-70	:	zeta chain associated protien kinase 70

List of Figures

List of Tables

INTRODUCTION

T cells and cell mediated immunity constitute one of two major cellular components of the adaptive immune response. The function of T cells is to recognize specific "non-self" antigens, during a process known as antigen presentation, leading to generation of specific responses that are tailored to maximally eliminate specific pathogens or pathogen-infected cells (Ballow, 2014).

Measurement of cell-mediated immunity can be undertaken by both in vitro and in vivo methods. It is, however, more problematic than humoral assessment as assays are plagued by difficulties in standardisation, biological variability, imprecision and technical complexity. Most tests are highly specialised and referral to a clinical immunologist is recommended (Limaye, 2010).

Delayed-type hypersensitivity skin testing provides a functional in vivo assessment of cellular immunity. The skin response following intradermal inoculation of antigen is dependent on antigen-specific memory T cells and results in local inflammation after 48–72 hours due to the recruitment of mononuclear cells (lymphocytes, monocytes) and neutrophils

(Limaye, 2010). Both standardized and non standardized candida intradermal skin tests are used in clinical practice with variable results (Ohri and Omaha, 2004; Yeo et al., 2009).

AIM OF THE WORK

This study is aimed to investigate the reliability of candida skin test in evaluation of T cell function in comparison to in-vitro assessment of T cell function in a group of healthy Egyptian infants using the manually prepared Candida intra-dermal skin test.

The ultimate objective is to validate the use of this test in the screening for T cell function in this age group.

REVIEW OF LITERATURE

CHAPTER 1
THE IMMUNE SYSTEM

The immune system is a complex network of cells and organs that functions to protect the body against pathogens. Any invader to the body will be attacked by the components of the innate immune system and, afterwards, by the specific immune system.Innate and specific immunities work conjointly (Protonotariou et al., 2010).

1.1 The innate immune system:

The innate immune system provides a rapid first line of defense, giving the adaptive immune system time to build up a more specific response. Components include phagocytic cells (neutrophils and monocytes in blood, macrophages and dendritic cells in tissues), antigen-presenting cells (APC), natural killer (NK) cells, polymorphonuclear leukocytes (PNL) (Parkin and Cohen, 2001).