

Assessment of Decidual Natural Killer Cells CD56⁺ Population in Placental Bed in Fetal Growth Restriction

Thesis

*Submitted for Partial Fulfillment of Master Degree in
Obstetrics and Gynecology*

By

Noha Mahmoud Kasim

M.B.B.Ch, December 2010

Faculty of Medicine- Ain Shams University

*Resident of Obstetrics and Gynecology El-sheikh Zayed Al-
Nahian Hospital*

Under Supervision of

Prof. Mohammad Alaa Mohi El-Din El-Ghannam

*Professor of Obstetrics and Gynecology
Faculty of Medicine, Ain-Shams University*

Prof. Mohamed ElMandooh Mohamed

*Professor of Obstetrics and Gynecology
Faculty of Medicine Ain-Shams University*

Dr. Mohammed Saeed El-Din El-safty

*Assistant professor of Obstetrics and Gynecology
Faculty of Medicine Ain-Shams University*

**Faculty of Medicine
Ain Shams University
2017**



Acknowledgment

- ✍ Firstly, thanks to **ALLAH**, Who gave me the power to finish this work.
- ✍ I'd like to express my sincere gratitude to my advisor **Prof. Mohammad Alaa Mohi El-Din El-Ghannam** Professor of Obstetrics and Gynecology, Faculty of Medicine, Ain-Shams University for the continuous support of my research.
- ✍ I am greatly honored to express my deepest thanks, gratitude and respect to **Dr. Mohamed ElMandooh Mohamed** professor of Obstetrics and Gynecology, Faculty of Medicine, Ain-Shams University for his guidance, supervision and continuous support.
- ✍ I am deeply indebted to **Dr. Mohammed Saeed El-Din El-safty** Assistant professor of Obstetrics and Gynecology, faculty of medicine Ain-Shams University for her guidance and unlimited assistance throughout this work.
- ✍ I'd like to thank **Dr.Nahla Mohamed Awad** consultant of Pathology in Early cancer Detection Unite at Ain Shams University Hospitals, for her patience, motivation and immense knowledge.
- ✍ Last but not least, I would like to thank my family for supporting me spiritually throughout writing this thesis and my life in general.

Noha Mahmoud Kasim

Contents

Subjects	Page
List of abbreviation	I
List of tables	II
List of figures.....	III
Introduction.....	1
Aim of the work.....	6
Review of literature	
- Chapter (1): Intrauterine Growth Restriction	7
- Chapter (2): Natural Killer Cell	36
- Chapter (3): Immunobiologic Adaptation of Pregnancy.....	42
- Chapter (4): Normal Placentation.....	50
Patients and methods	64
Results	70
Discussion	93
Summary	99
Conclulsion	102
Recommendations.....	103
References.....	104
Arabic summary	

List of Abbreviations

AC	: Abdominal circumference
ACA	: Anticardiolipin antibodies
ACOG	: American Congress of Obstetricians and Gynecologists
AFI	: Amniotic fluid index
AIUM	: American Institute of Ultrasound in Medicine)
Ang	: Angiopoietin
APS	: Antiphospholipid syndrome
ART	: Artificial reproductive techniques
BPD	: Biparietal diameter
BPP	: Biophysical profile
CMV	: Cytomegalovirus
COPD	: Chronic Obstructive Pulmonary Disease
CRL	: Crown-rump length
CTG	: Cardiotocography
CXC R3	: Chemokine receptor R3
CXCR1	: Chemokine receptor R1
DCs	: Dendritic cells
DNA	: Deoxyribonucleic acid
dNK	: Decidual Natural killer cells
ECM	: Extracellular matrix
EFW	: Estimated fetal weight
EFW	: Estimated fetal weight
eNK	: Endometrial Natural killer cells
EVT	: Extravillous trophoblast
FcγRIII	: Fragment crystallizable

FHR	: Fetal heart rate
FL	: Femur length
GM-CSF	: Granulocyte–macrophage colony-stimulating
GRIT	: <i>The Growth Restriction Intervention Trial</i>
HC	: Head circumference
HIV	: Human immunodeficiency virus
HLA	: Human leukocyte antigen
HPF	: High Power Field
IFNγ	: Interferon- γ
IG	: Immunoglobulin
IL-2Rβ	: Interleukin-2 receptor subunit beta
IL-3	: Interleukin-3 receptor
IL-8	: Interleukin-8 receptor
IL-IRI	: Interleukin-iri
IP-10	: Inducible protein 10
IQ	: Intelligence quotient
IUGR	: Intrauterine growth restriction
IVF	: In vitro fertilization
LBW	: Low birth weight
MCA	: Middle cerebral artery
Mhc	: Major histocompatibility complex
Nk	: Natural killer T cells
NKT	: Natural killer cells
NO	: Nitric oxide
PGF	: Placental growth factor
PGs	: Prostaglandins
PIGF	: Phosphatidylinositol-glycan biosynthesis class F

	protein
PIGF	: Placental growth factor
RBC	: Red blood cell
RCOG	: Royal College of Obstetricians and Gynecologists
'SFH'	: Symphysis fundal hieght
sFlt1	: Soluble fms-like tyrosine kinase-1
sFlt-1	: Soluble fms-like tyrosine kinase receptor-1
SGA	: Small for gestational age
SLE	: Systemic lupus erythematosus
TB	: Tuberculosis
TCR	: T-cell receptor
TGF- β	: Transforming growth factor beta 1
Th type-1	: T helper cells type-1
Th2	: Type-2 T-helper
TNF-α	: Tumour necrosis factor- α
TORCH	: Toxoplasmosis, rubella, cytomegalovirus, herpes simplex
UA	: Umbilical artery
VEGF	: Vascular endothelial growth factor

List of Tables

Tables No.	Title	Page No.
1	Etiology of IUGR	18
2	Perinatal and pediatric complications from IUGR	24
3	Approximate proportions of NK cells in blood and deciduas	46
4	Comparison between cases and controls as regard Age, Gestational Age and BMI	71
5	Comparison between cases and controls as regard Parity, Previous Cs and Previous Abortion	75
6	Comparison between cases and controls as regard Systolic, Diastolic B.P and HB	78
7	Comparison between cases and controls as regard Birth weight and Apgar score at 5 minutes	82
8	Comparison between cases and controls as regard Immunohistochemical scores	85
9	Comparison between cases and controls as regard dNK Cell Density	87

List of Figures

Figures No.	Title	Page No.
1	Symphysis fundal hieght 'SFH'	26
2	NK cell changes During the adaptive process of endometrium to decidua	39
3	The blood flow through the maternal uterine arteries is increased by the infiltration of the arterial media and endothelium by extravillous trophoblast cells	54
4	Scheme of placental circulation	57
5	The diagram depicts the 2 different pathways leading to preeclampsia or IUGR	60
6	The diagram represents the early development of the trophoblast lineage	62
7	Flow chart shows the study groups.	70
8	Comparison between cases and controls as regard Age.	71
9	Comparison between cases and controls as regard Gestational Age.	72
10	Comparison between cases and controls as regard BMI.	73

Figures No.	Title	Page No.
11	Comparison between cases and controls as regard Parity	75
12	Comparison between cases and controls as regard previous CS	76
13	Comparison between cases and controls as regard previous abortion	77
14	Comparison between cases and controls as regard Systolic B.P.	79
15	Comparison between cases and controls as regard Diastolic B.P.	80
16	Comparison between cases and controls as regard Mean Hb.	81
17	Comparison between cases and controls as regard Birth weight	83
18	Comparison between cases and controls as regard Apgar score.	84
19	Comparison between cases and controls as regard Immunohistochemical scores	86
20	Comparison between cases and controls as regard dNK Cell Density.	88
21	dNK cells CD56 in subject from control group shows high density by immunohistochemical stain in HPF x100	89

Figures No.	Title	Page No.
22	. dNK cells CD56 in subject from control group shows high density immunohistochemical score 3+ in HPF x400	89
23	dNK cells CD56 in subject from control group shows high density by immunohistochemical stain in HPF x100.	90
24	dNK cells CD56in subject from control group shows high density by immunohistochemical score 4+ in HPF x400.	90
25	dNK cells CD56in patient complicated IUGR shows low density by immunohistochemical stain in HPF x100	91
26	dNK cells CD56 in patient complicated IUGR shows low density by immunohistochemical score 2+ in HPF x400	91
27	dNK cells CD56in patient complicated IUGR shows low density by immunohistochemical stain in HPF x100.	92

Figures No.	Title	Page No.
28	dNK cells CD56in patient complicated IUGR shows low density by immunohistochemical stain x100.	92

Introduction

Several definitions and terminology has been used to define IUGR, including but not limited to estimated fetal weight <25 %, <15 %, <10 %, <5 %, <3 %, <2.5 %, and <1 % for gestational age. Other definitions of IUGR include estimated weight less than 2 standard deviations below the mean weight for gestational age (*Suhag and Berghella, 2013*).

The causes of IUGR are broadly described under three main categories: maternal, fetal, and placental (*Hendrix and Berghell, 2008*).

Normal fetal growth reflects the interaction between the genetically predetermined growth potential and the presence of a healthy fetus, placenta and mother (*Nardozza et al., 2017*).

A Several maternal demographic factors have been associated with IUGR. Women at extremes of reproductive age, especially young maternal age, are at increased risk for IUGR. There no association between maternal age and low birth weight and reported an independent effect of social factors, such as ethnicity, poverty status, age at menarche, maternal height, net maternal weight gain, and smoking during pregnancy, on birth weight in adolescent mothers (*Suhag and Berghella, 2013*).

Fetal factors can vary from genetic causes, congenital malformations, fetal infection, or other causes, including multiple pregnancies. Genetic causes can contribute to 5-20 % of IUGR, especially for early onset growth restricted fetuses. Genetic causes further include various abnormalities, such as chromosomal abnormalities, e.g., trisomy 21, 18, 13, and 16. Of these, trisomy 18 is associated with more severe IUGR compared with trisomy 21 or 13. Trisomy 16 is known to be a lethal chromosomal abnormality in the non-mosaic state; however, in the presence of placenta mosaicism, trisomy 16 can result in IUGR (*Baschat et al., 2012*).

Placental insufficiency accounts for many cases of IUGR and can affect up to 3 % or more of all pregnancies. The pathogenesis of IUGR is not well defined; defects in the placental circulation and transport can affect the nutrient transport to the fetus, resulting in IUGR. The relative decrease in placental mass and function can result in the development of IUGR. Several animal models have shown that fetal growth can be impaired when up to 50 % of the placental mass is removed. Like the animal model, growth-restricted human fetuses are noted to have approximately a 24 % smaller placental weight compared with a normally grown fetus (*Brett et al., 2014*).

The asymmetrical IUGR fetuses are noted to be at higher risk for major anomalies, low birth weight, perinatal

mortality, preterm delivery, cesarean section, and overall poor outcomes, compared to symmetrical IUGR (**Dashe et al., 2000**). Umbilical artery Doppler studies and antenatal surveillance are very good predictors of pregnancy outcomes in all types of IUGR (**Alfirevic and Gyte, 2010**).

Perinatal morbidity and mortality is significantly increased in the presence of birth weight less than 10th percentile. After prematurity, IUGR is the second leading cause of perinatal mortality. IUGR fetuses have approximately a fivefold to tenfold increased risk of dying in utero, with up to 23 % to 65 % of stillbirths (**Chen et al., 2011**).

The first step in diagnosis of fetus with growth restriction is to establish accurate dating. There are several formulae to date the pregnancy from early biometrics, with low systematic and random errors. Crown-rump length (CRL) is a biometric parameter that can be measured in the early stages of gestation. Technically the main limitation is the progressive bending of the embryo which makes measurements less reliable beyond 12-13 weeks of gestation (or 60-70 mm). If possible, below the 14 weeks, all obstetric ultrasound units are currently recommended to adopt this method of assessing gestational age from crown rump length. From then on, it seems conceptually more appropriate to use cephalic (head circumference) and/ or femur (femur length) biometrics (**Xu et al., 2015**).