A STUDY OF ASSOCIATION OF HIGH SERUM PROLACTIN LEVELS WITH OTHER BIOCHEMICAL PARAMETERS IN PRE ECLAMPSIA

Dissertation submitted to

THE TAMILNADU DR. M.G.R MEDICAL UNIVERSITY,

CHENNAI – 600032

In partial fulfilment of the requirement for the degree of Doctor of Medicine in Biochemistry (Branch XIII)

M.D. (BIOCHEMISTRY)

MAY 2020

DEPARTMENT OF BIOCHEMISTRY

COIMBATORE MEDICAL COLLEGE

COIMBATORE – 14.

UNIVERSITY REGISTRATION NO. 201723651

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Certificate

CERTIFICATE

This dissertation entitled **"A STUDY OF ASSOCIATION OF HIGH SERUM PROLACTIN LEVELS WITH OTHER BIOCHEMICAL PARAMETERS IN PRE ECLAMPSIA"** is submitted to The Tamil Nadu Dr.M.G.R Medical University, Chennai, in partial fulfilment of regulations for the award of M.D. Degree in Biochemistry in the examinations to be held during May 2020.

This dissertation is a record of fresh work done by the candidate **Dr**.**T.DANIA TAMILSELVI,** during the course of the study (2017 – 2020). This work was carried out by the candidate herself under my supervision.

GUIDE:

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CERTIFICATE II

This is to certify that this dissertation work titled "**A STUDY OF ASSOCIATION OF HIGH SERUM PROLACTIN LEVELS WITH OTHER BIOCHEMICAL PARAMETERS IN PRE ECLAMPSIA**" of the candidate **Dr.T.DANIA TAMILSELVI** with registration Number 201723651 for the award of **M.D. DEGREE** in the branch of **BIOCHEMISTRY**. I personally verified the urkund.com website for the purpose of plagiarism Check. I found that the uploaded thesis file contains from introduction to conclusion pages and result shows 3 (THREE) percentage of plagiarism in the dissertation.

Guide & Supervisor sign with Seal.

Declaration

DECLARATION

I, **Dr.T.DANIA TAMILSELVI,** solemnly declare that the dissertation entitled **"A STUDY OF ASSOCIATION OF HIGH SERUM PROLACTIN LEVELS WITH OTHER BIOCHEMICAL PARAMETERS IN PRE ECLAMPSIA"** was done by me at Coimbatore Medical College, during the period from March 2018 to March 2019 under the guidance and supervision of **Dr.S.MANIMEKALAI M.D.,** Professor and Head, Department of Biochemistry, Coimbatore Medical College, Coimbatore.

This dissertation is submitted to The Tamilnadu Dr. M.G.R. Medical University towards the partial fulfilment of the requirement for the award of M.D. Degree (Branch - XIII) in Biochemistry. I have not submitted this dissertation on any previous occasion to any University for the award of any degree.

Place: Coimbatore **Dr.T.DANIA TAMILSELVI**

Date:

Acknowledgement

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"Gratitude is the humble gift , I can give to my beloved Teachers".

I wish to express my sincere thanks to our respected **Dean Dr.B.ASOKAN,M.S,MCh,** for having allowed me to conduct this study in our hospital.I express my heart felt thanks and deep gratitude to the Head of department of Biochemistry Prof **Dr S.MANIMEKALAI M.D.,**for her generous help and guidance in the course of the study.

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I thank my patients who formed the back bone of this study without whom this study would have not been possible.Last but not the least **I** thank my parents, husband and kids for having extended unconditional support throughout my life.

Abbreviations

ABBREVIATIONS

Flt-1	fms like tyrosine kinase
Flk-1	kinase insert domain receptor
VEGFR 1,2,3	Vascular endothelial growth factor receptor 1,2,3.
PlGF	Placental growth factor
VEGF	Vascular endothelial growth factor
sFlt-1	Soluble fms like tyrosine kinase.
SNP	Single nucleotide polymorphism.
NLRP	Nod like receptors with a pyrin domain.
ENG	Endoglin
ALK-5	Activin like kinase
HO-1	Heme oxygenase
ET	Endothelin-1
AT1-AA	Angiotensin II receptor 1 autoantibodies.
uNK	uterine Natural killer cell
DC	Dendritic cell
MMPs	Matrix metalloproteinase.
Treg	Regulatory T cells.

RAS	Renin Angiotensin system
ACE	Angiotensin converting enzyme
AT-1	Angiotensin-1
PAI-1	Plasminogen activator inhibitor.
VSM	Vascular smooth muscle.
STBM	Syncytiotrophoblast microparticles.
HSP-70	Heat shock protein-70
HMGB-1	High mobility group box-1
PP13	Placental Protein 13
PRLR	Prolactin receptor
TH mRNA	Tyrosine hydroxylase messenger Ribonucleic Acid.
ng/ml	nanogram per millilitre.
LDH	Lactate dehydrogenase.
AST	Aspartate aminotransferase
ALT	Alanine aminotransferase.

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Introduction

INTRODUCTION

"Pregnancy is nature's precious boon which has to be nurtured during its entire nine months, to achieve good maternal and fetal outcome"

"**What makes the blood pressure in pregnancy to rise**

Is still a mystery to many a wise?

How can we find a method of cure

When the causative factor still remains obscure"

Pre eclampsia is a pregnancy related multisystem disorder, which is the leading cause of maternal and fetal mortality and morbidity. It can clinically manifest after 20 weeks of gestational age with

- Hypertension, Blood pressure > 140/90 mm of Hg.
- Protein in the urine.

This usually resolves within 42 days after delivery.

It is one of the extensively studied disease, yet its etiopathogenesis remains unclear. It is called as the "Disease of theories".

Among the various hypotheses, one is about the prolactin fragments and its antiangiogenic property.

In pre eclampsia, the placenta is abnormal and characterized by poor trophoblastic invasion. Pre eclampsia upregulates Trophoblastic cathepsin D, which cleaves 23 K Da Prolactin into its fragments namely 14 K Da and 16 K Da,both exhibits anti angiogenic factors. Its majority form, 16 K Da blocks Vascular Endothelial Growth Factor (VEGF) and Placental Growth Factor (PlGF). It is thought that this results in hypoxia, oxidative stress and the release of factors that promote endothelial dysfunction, inflammation, and other possible reactions.

Aims and Objectives

AIM OF THE STUDY

To find the role of serum prolactin and other biochemical parameters such as serum uric acid, lactate dehydrogenase, alkaline phosphatase and 24 hours urine protein in pre eclampsia.

OBJECTIVES:

- To estimate the level of serum prolactin, serum uric acid, serum lactate dehydrogenase, serum alkaline phosphatase and 24 hours urine protein in Pre eclampsia patients.
- To evaluate the correlation of elevated levels of serum Prolactin and other biochemical parameters in Pre eclampsia patients in a tertiary care center.

Review of Literature

REVIEW OF LITERATURE:

HISTORY OF PRE ECLAMPSIA:

What is known as Pre eclampsia a millennium later, was first described in 400 BC by Hippocrates.

Pre eclampsia was not considered as a disease in 4th century, when Hippocrates suggested an entity of unhealthy pregnancy accompanied by headache, heaviness (fluid overload) and convulsions.

He postulated a theory that human body was made of four humors namely

- Blood
- Phlegm
- Black bile
- Yellow bile

And the illness was caused due to the imbalance between these humors[(1)].

Hence treatment was based on the myth that a woman needed to be pregnant, or lactating or menstruating regularly in order to eliminate the excess of body fluids.

During pregnancy associated pre eclampsia, various remedies were followed to restore fluid balance like purging, alterations in diet and letting out of human blood.

Since then until late 19th century, there was limited progress in understanding Pre eclampsia and eclampsia.

Later the specific Pre eclampsia and eclampsia syndrome was delineated and classified by various medical pioneers, which are quoted as below.

- Bossier de Sauvages in 1739 differentiated epilepsy from eclampsia.
- Demanet in 1797 discovered extreme generalized swelling in eclampsia.
- Pierre Rayer in 1840 recognized proteins in urine.
- John Lever in 1843 proved that proteinuria is exclusive to Pre eclampsia and not related to kidney disorders.
- Scipione Riva Rocci in 1896 discovered mercury manometer to measure blood pressure.(1)

According to Chesley it was Galeni in 1829 who coined the term eclampsia (ΕxλαμΨιεζ) in Greek meaning lightning, which perhaps relates to how unexpectedly, suddenly a pregnant women throws convulsions in eclampsia.[2]

In 20th century, various theories on disease causation was put forward by researchers across the world, which will be discussed in detail in etiopathogenesis.

INCIDENCE AND PREVALENCE:

Reduction of maternal mortality has been a long-term goal to be reached in global health policy. In order to achieve this, we need to understand the causes of maternal mortality as summarized in figure 1.

Using International Classification of Diseases definition of maternal mortality and analyzing various regional and global estimate from 2003-2009, the following data was collected and depicted in figure 2.

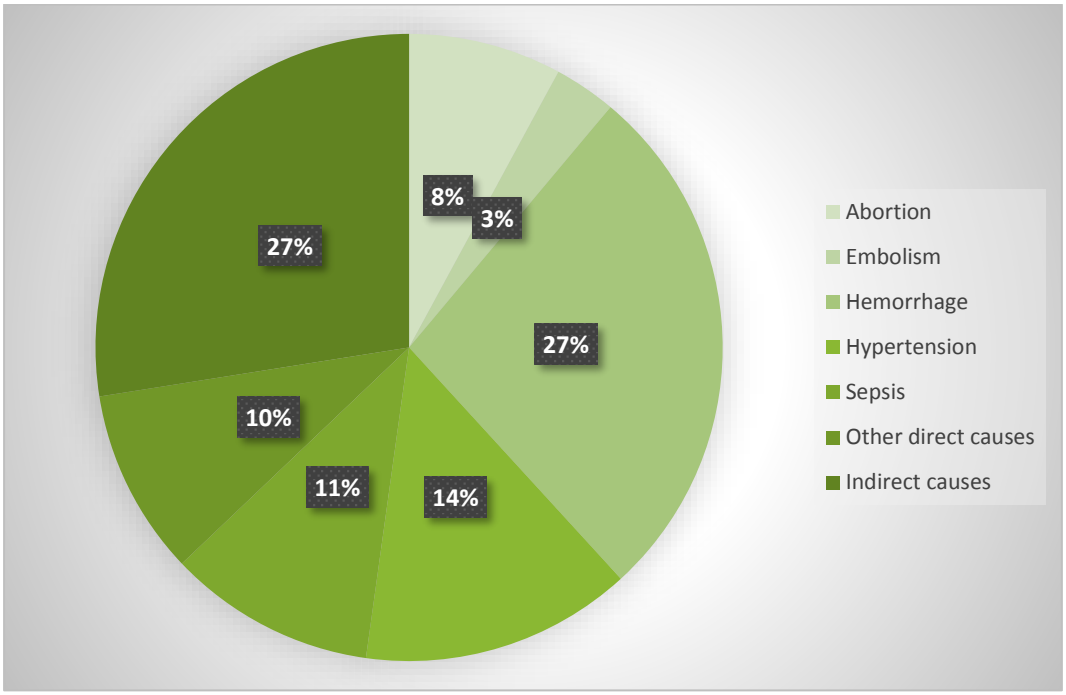
TABLE 1

DISTRIBUTION OF CAUSES OF MATERNAL MORTALITY[4]

Causes	South Asia	World wide
Abortion	5.9 %	7.9 %
Embolism	2.2 %	3.2 %
Hemorrhage	30.3 %	27.1 %
Hypertension	10.3 %	14.0 %
Sepsis	13.7 %	10.7 %
Other direct causes	8.3 %	9.6 %
Indirect causes	29.3 %	27.5 %

FIGURE 1: CAUSES OF MATERNAL MORTALITY

IN THE WORLD



Including sub categoriesof

- Other Direct causes:

  Complications occurring during delivery.

  Obstructed labor.

- Indirect causes:

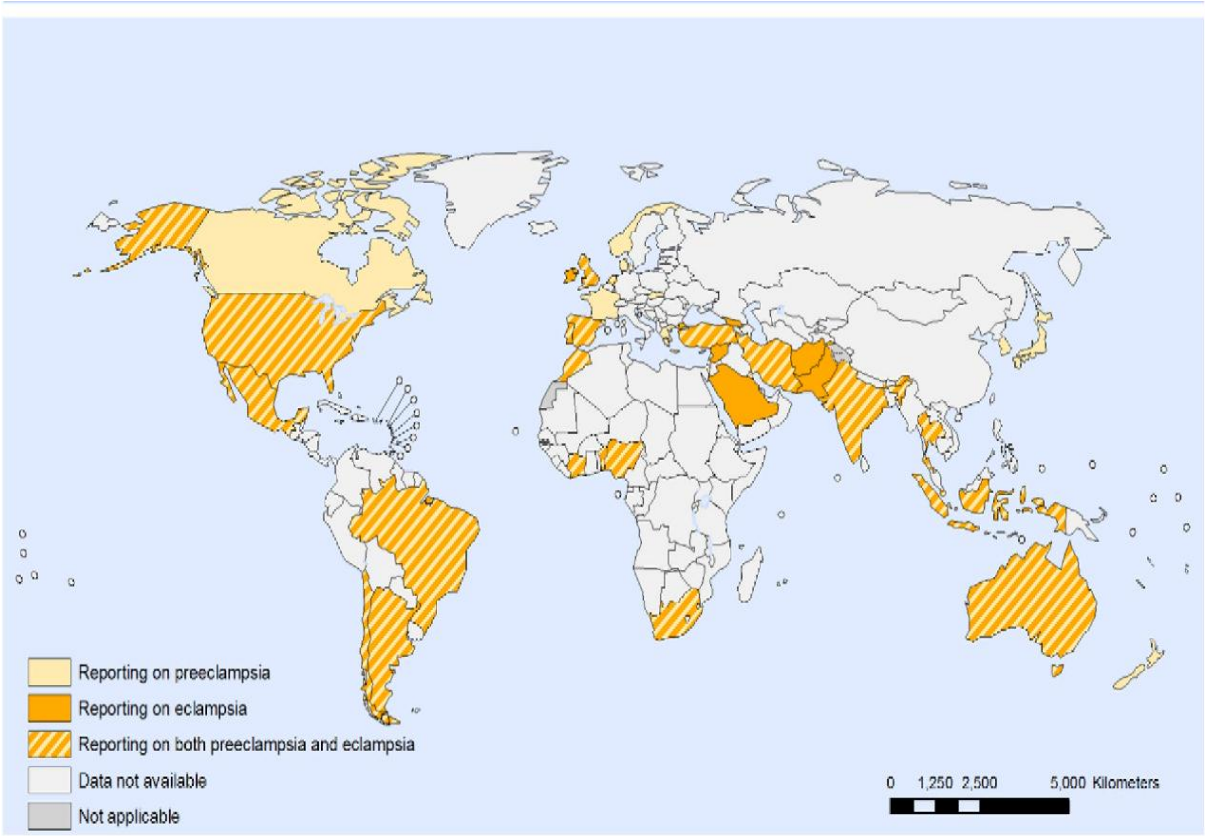
  Medical disorders

  HIV related maternal deaths.

Pre eclampsia affects about 5-8% of all pregnancies throughout the world, and is responsible for 60,000 maternal deaths and 5 lakh premature births every year.[3]

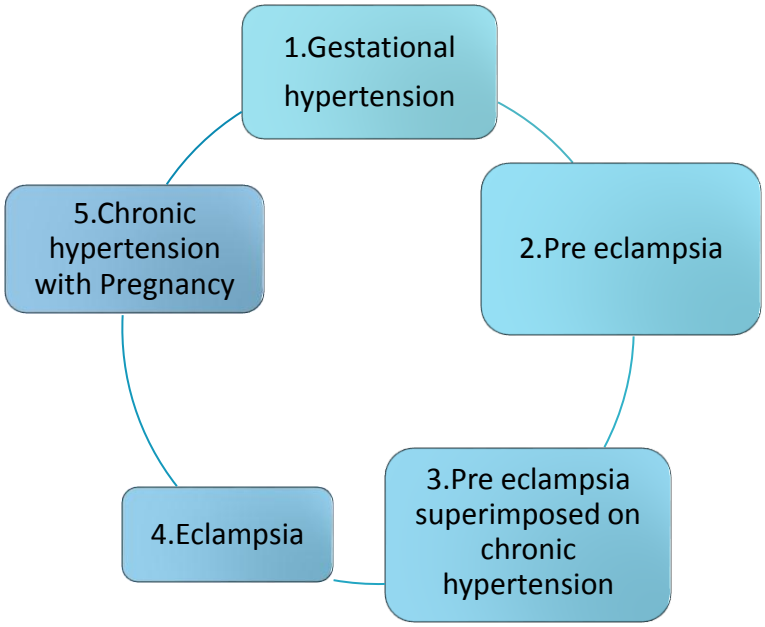
FIGURE 2:

WORLD MAP SHOWING PREVALENCE OF PRE ECLAMPSIA



CLASSIFICATION OF HYPERTENSIVE DISORDERS IN PREGNANCY[4]

According to NHBPEP – National High Blood Pressure Education Program, the classification is as follows:



1.Diagnosis of Gestational Hypertension:

- A blood pressure of ≥140/90 mm of Hg for the first time in pregnancy.
- Absence of Protein in Urine.
- Blood pressure returns to normal within 42 days.

2. Diagnosis of Pre eclampsia:

- A new onset of blood pressure of ≥140/90 mm of Hg after 20 weeks of pregnancy.
- Excretion of protein in the urine (>300 mg for 24hrs urine sample).

3. Diagnosis of Pre eclampsia superimposed on chronic hypertension:

- ❖ A woman with hypertension who had no protein excretion in urine, suddenly develops Proteinuria after 20 weeks of gestation.
- ❖ Development of thrombocytopenia (Platelet count < 1,00,000/mm^3) in a woman with proteinuria and hypertension before 20 weeks of gestation.

4. Diagnosis of Eclampsia:

- ❖ In a Pre eclampsia woman, sudden onset of generalized tonic and clonic seizures or coma during pregnancy or in postpartum, when other causes of cerebral condition are ruled out.

5.Diagnosis of Chronic Hypertension:

- ❖ A blood pressure of $\geq$140/90 mm of Hg before pregnancy.
- ❖ Persists even after 42 days of delivery.
- ❖ When secondary causes of hypertension and hydatid mole are ruled out.

RISK FACTORS:

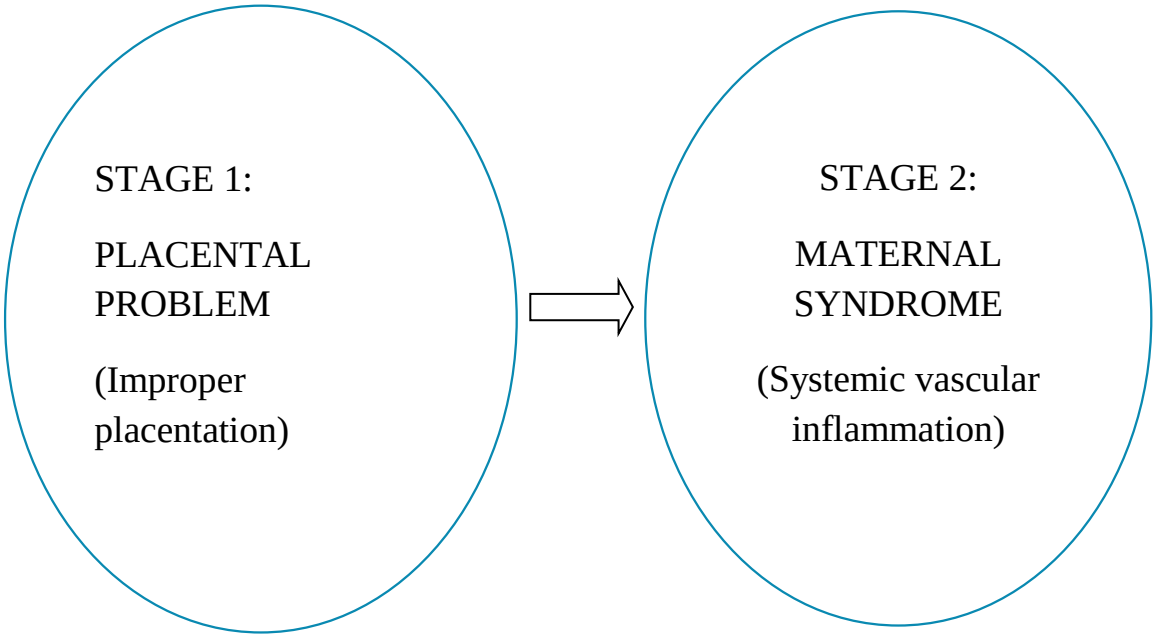
TABLE 2: List of Various risk factors for developing Pre eclampsia.[5]

Major Risk Factors
Pre eclampsia in Previous pregnancy.
Chronic Hypertension.
Gestational Hypertension.
Multiple Gestation.
Obesity (BMI>30) complicating Pregnancy.
Antiphospholipid Syndrome.
Moderate Risk Factors
Systemic Lupus Erythematosus.
Previous History of Stillbirth.
Nulliparity.
Chronic Kidney Disease.
Advanced Maternal Age > 35.
Genetic susceptibility of mother.
Rare Risk Factors
Family History of Pre eclampsia.
Conception of Trisomy 13 Fetus.

ETIOPATHOGENESIS

Despite extensive research on Pre eclampsia for the last few decades, it is labelled “disease of theories” as its exact etiology is unclear.

Traditional theory states that, Pre eclampsia develops in two stages. [6]



STAGE 1 OF PRE ECLAMPSIA: IMPROPER PLACENTATION.

Implantation occurs

↓

Until 8 weeks of gestation, cytotrophoblast invades spiral arteries and plugs it.

↓

Opposite to cord insertion, unplugging starts which continues circumferentially around the chorionic sac.

↓

Due to oxidative stress, around 8-9 weeks, chorionic villi get atrophied to form chorionic laeve.

↓

Around 10 – 12 weeks, placenta is formed from the remaining mature villi, which were able to withstand the oxidative stress.

↓

Spiral artery remodeling occurs completely when it is invaded by trophoblast deep into myometrial segments.

↓

Failure of this remodeling results in pre eclampsia, causing pulsatile, high pressure uteroplacental perfusion ultimately damaging chorionic villi.

↓

Factors released by these damaged chorionic villi like syncytiotrophoblast microvesicles, VEGF, PlGF etc, cause maternal syndrome of pre eclampsia.

STAGE II – MATERNAL SYNDROME:

Microvillous epithelium of placenta is in contact with maternal blood

These are lined with syncytiotrophoblast.

Oxidative stress causes dysfunction of syncytiotrophoblast which stimulates the release of various factors.

These factors can be Proinflammatory, Antiangiogenic (sFlt-1).

Ultimately results in systemic inflammatory response in maternal side.

Various schools of thought have proposed a few pathogenic mechanisms due to the following factors:

I. Genetic factor

II. Abnormal Placental vasculogenesis

III.Placental Angiogenic factors

IV. Excessive Oxidative stress

V. Maladaptation of Immune system

VI. Renin-Aldosterone- Angiotensin II

VII.Synciotrophoblast debris

I.GENETIC FACTOR:

Clustering of Pre eclampsia within families and a few ethnic groups have been noticed since 19th century. This has suggested researchers to explore the role of genetic involvement in this disorder. It is not an easy task to decipher the genetic involvement in pre eclampsia. The challenges encountered are

- As it is a disorder of pregnancy, two genotypes have to be considered, namely both mother and unborn fetus.
- It is difficult to delineate Pre eclampsia from that of Pre eclampsia superimposed on chronic hypertension, non-proteinuric gestational hypertension, due to sliding scale of severity.

So, it is necessary to assess both maternal and fetal genotype, with more focus laid on former genotype.

However recent researches strongly indicate the partner's role in causation of Pre eclampsia.(6)

- Decreased duration of sperm exposure.
- Pregnancy due to donated gametes.
- Partner's HLA typing.

All the above are postulated to be the causative factor for the development of Pre eclampsia in a woman married to so called dangerous partner. (6)

Pre eclampsia is a polygenic disorder, which involves several genes in different signaling pathways. It is not the genetic variants alone, but also the environmental factors, gene-gene interaction (Epistasis), epigenetic modification which plays a key role in the development of this disorder.

Various candidate genes studied have been segregated under the groups, based on the following pathological mechanisms:

- Thrombophilia.
- Endothelial function.
- Oxidative stress.
- Vasoactive protein.
- Immunogenetics.
- Lipid metabolism.

In this context, evaluation of candidate genes would help to identify high risk mother at an earlier stage for better management of complications and follow up.

Genome wide association studies (GWAS) have showed around 70 biological candidate genes involved in pre eclampsia.

Among these, here is a list of few candidate genes involved in Pre eclampsia in table 3.

TABLE 3:

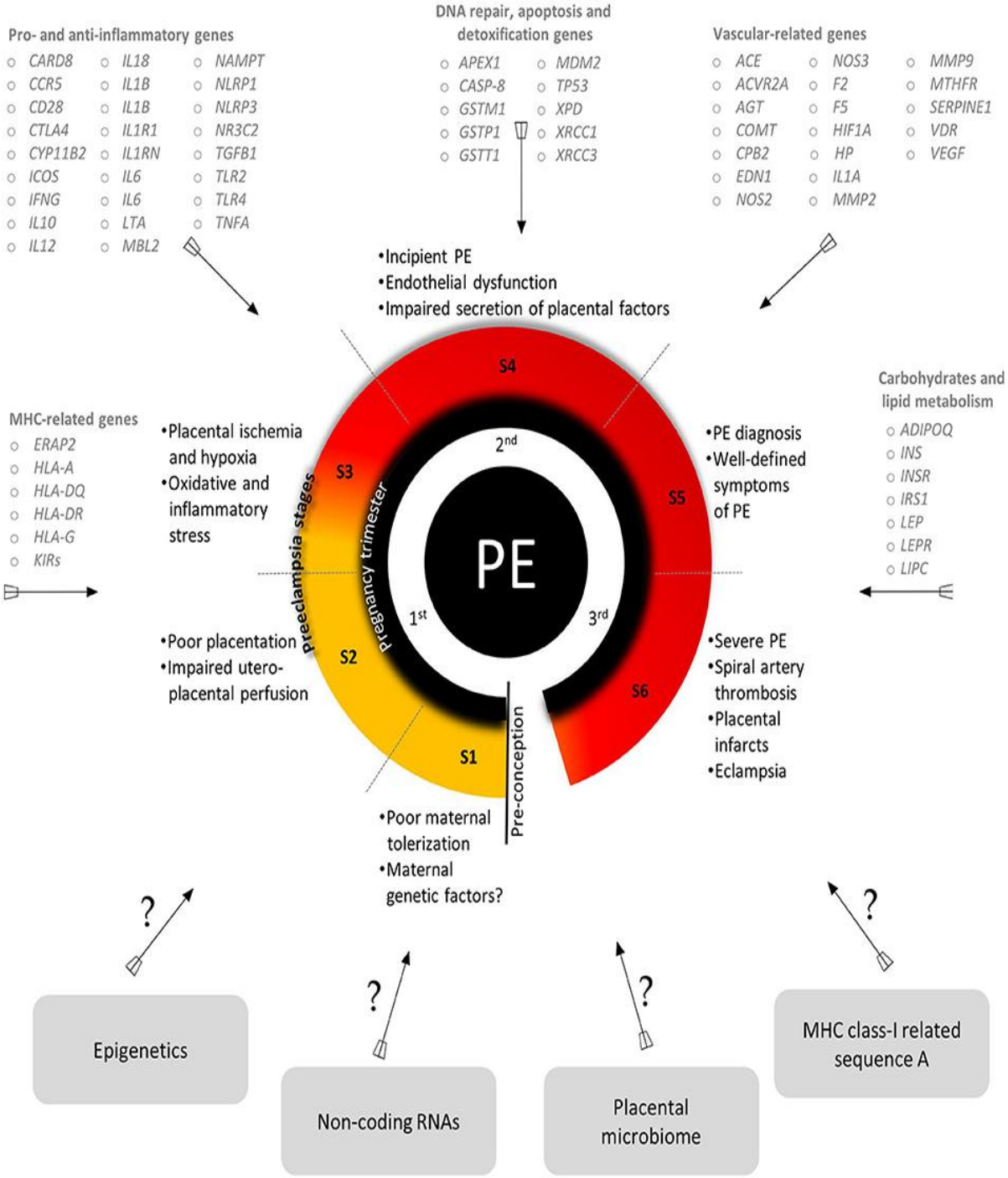
PRE ECLAMPSIA-CANDIDATE GENES.[7]

Underlying pathophysiology	Gene name	Common polymorphism investigated
Thrombophilia	Prothrombin Factor V Leiden Methylenetetrahydrofolate Integrin glycoprotein IIIa Plasminogen activator factor – 1	G20210A 506Gln>Arg C667T C98T I/D promotor
Endothelial function	Vascular endothelial growth factor receptor-1 Endothelial nitric oxide synthase-3 Vascular endothelial growth factor	TG repeat 298Glu>Asp C936T
Oxidative stress and Lipid metabolism	Apolipoprotein E Glutathione S-transferase Microsomal epoxide hydrolase	C866T A313G 113Tyr>His
Vasoactive proteins	Angiotensinogen Angiotensin converting enzyme	235Met>Thr I/D intron 16

Apart from these candidate genes, recent studies have been focused on other susceptible genes which are compiled under the following figure 3.

FIGURE 3:

AN INTEGRATED PICTURE OF KEY EVENTS IN PRE ECLAMPSIA.[8]



GENETIC ASPECT OF PRO AND ANTI-INFLAMMATORY MEDIATORS:

Pendeloski et al., conducted a study in Brazilian women and found an inverse association between single nucleotide polymorphism of ICOS(-1564 T/C) and Pre eclampsia.

Aguilar et al., suggested genotype +869TT as a protective factor against pre eclampsia.

De Lima et al., proved the association of SNP IFNA (-308), IL10 (-1082), TGFB1(+10;25), IL6(-174), IFNG(+874) with pre eclampsia.

Leme Galvao et al., showed the association of “rs1143630T” allele with pre eclampsia.

Pinheiro et al., observed that levels of IFN-γ and IL-6 were increased in Pre eclampsia and have a positive correlation with IFNG+874T allele.

Turner et al., evidenced the association of Mannose -binding lectin (MBL) with polymorphism of MBL2 gene in exon 1at codons 57 (allele C,rs18000451), 54(allele B,rs1800450).

Pontillo et al., evidenced the association of SNP of NLRP1 rs12150550 with pre eclampsia.

To summarize these are the various polymorphism observed in genes involved in inflammatory mediators.

GENETIC ASPECT OF VASCULAR MEDIATORS:

Nitric oxide plays an important role as a regulatory factor in ovulation, implantation, maintenance of gestation, delivery etc.

Pre eclampsia is postulated to be due to imbalance in nitric oxide levels, due to SNP in inducible and endothelial nitric oxide synthase genes.[8]

Serrano et al., Sandrim et al., Diaz Olguin et al., Leonardo et al., Alpoim et al., Muniz et al., found the association of eNOS (-786T, intron-4 b/a, Glu298Asp) with Pre eclampsia risk.

Chedraul et al., Canto et al., evidenced the link between MTHFR (C677T) with risk of pre eclampsia.

Sandrim et al., Cunha et al., associated VEGF(C936T,C -2578A) and VEGF (G634C) as a protective factor pre eclampsia.

Luizon et al., confirmed the association of protection factor for Pre eclampsia namely eNOS (T786C), MMP9 (C1562T).

Ferrera et al., found the association of ACVR2A (rs1424954, rs1014064, rs3768687, rs2161983, rs142941) with risk of severe onset of pre eclampsia.

Hill et al., found the association of COMT (rs6269.rs4680, rs4633, rs4818) and MTHFR (C6771) with Pre eclampsia risk.

GENETIC VARIANTS OF HISTOCOMPATIBILITY:

Kovats et al., elucidated the role of HLA-G gene expression in human trophoblast cells.

HLA-G interacts with decidual macrophages, dCD4+,dCD8+,dNK and inhibits or activates various immunological functions.

Carreiras et al., found HLA-A, HLA-G, HLA-DRB1, DQB1, DQA1 alleles as a risk for developing pre eclampsia.

GENETIC ASPECT OF METABOLIC SYNDROME IN PRE ECLAMPSIA:

As a normal adaptation to gestation, changes occur in the metabolism of carbohydrate and lipid in the body. Insulin sensitivity decreases as period of gestation increases. Hyperglycemia in pregnancy is related to many adverse outcomes in both the fetus and mother like pre eclampsia. Genetic aspects of various critical mediators like Adiponectin, hepatic lipase, leptin is discussed here.

Machado et al., found the association of ADIPOQ (11391G>A, 45T>G, 11377C, 276G>T) with pre eclampsia.

Farias et al., associated LEP (D2548A), LEPR (Lys109Arg, Gln223Arg) in both gestational hypertension and pre eclampsia.

Enquobahrie et al., evidenced the link between LIPC(-514C>T) in overweight pregnant mothers and pre eclampsia.[9]

GENETIC VARIANTS IN DNA REPAIR, APOPTOSIS, DETOXIFICATION:

Oxidative stress plays a major role in endothelial dysfunction also causes peroxidation of lipid membrane and DNA damage. Pre eclampsia is caused by SNP of genes coding for apex nuclease, Glutathione-S-transferase etc.

Canto et al., associated GSTP1(313A>G) as a protective factor in pre eclampsia.

Sandoval carrillo et al., linked GSTM1 deletion and GSTM1/ GST11 deletion with pre eclampsia.

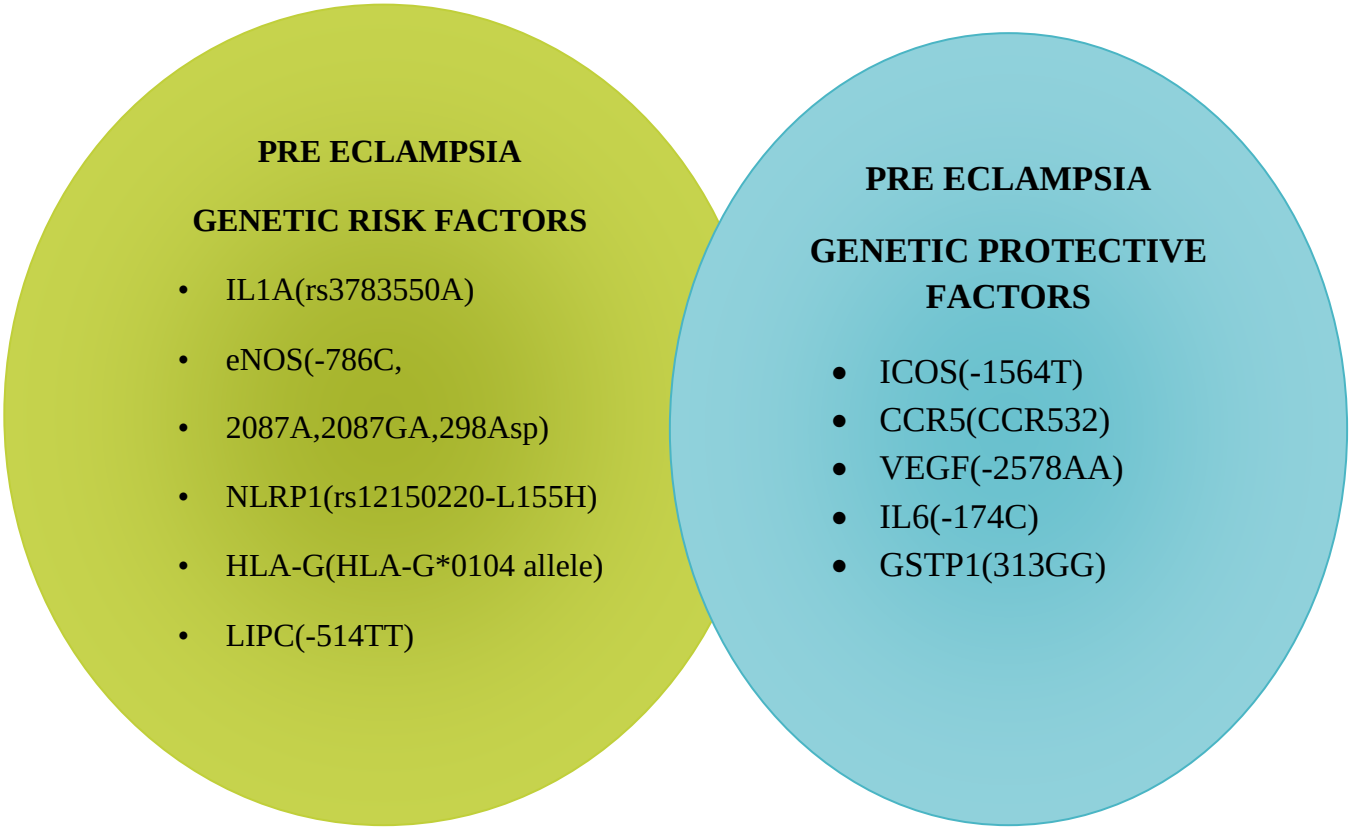
He also found the association of APEX1 (Asp148Glu), XRCC (Arg399Gln), XPD (Lys751Gln), XRCC3 (Thr241Met) with pre eclampsia. In particular APEX1 148Glu allele is evidenced by severe disorder of pre eclampsia.

Orlando et al., studied genetic variants like CASPASE-8 in apoptosis gene in Brazilian mothers and found no association with pre eclampsia.

Despite innumerous studies in understanding the role of maternal, paternal, fetal and placenta in etiopathogenesis of pre eclampsia, a predictive biomarker still remains elusive. These challenges can be overcome when in future, studies are centered towards elucidating the molecular basis of pre eclampsia. Figure 4 summarizes the various gene polymorphism

FIGURE 4:

SUMMARY OF POLYMORPHISM VARIANTS FOUND IN BOTH RISK AND PROTECTIVE FACTORS IN PRE ECLAMPSIA:



II.ABNORMAL PLACENTAL VASCULOGENESIS:

Development of placenta plays a central key role in Pre eclampsia and recent studies have scrutinized how the vascular remodeling in placenta contributes to this.

In normal pregnancy, two cardiovascular changes occur to provide a good effective blood supply to the fetus:

1. Blood from the lower limbs are diverted away to uterus.
2. Maternal cardiac output and blood volume is increased over 1/3rd, yet the maternal blood pressure drops.

This paradox is explained by profound reduction in maternal systemic vascular resistance by vascular remodeling. (10)

In pre eclampsia, placental ischemia and hypo perfusion is caused due to failure of vascular remodeling which is evidenced by pathological findings:

- Intimal thickening
- Deposition of fibrin
- Acute atherosis
- Endothelial damage
- Necrosis.

In Figure 5, anatomy of uterine and placental vasculature clearly depicts the changes that occur from non-gravid uterus to that in normal pregnancy, immediate post-partum and severe pre eclampsia.

In normal pregnancy, large arterio-venous shunts are seen.

In Immediate post-partum period, still arterio-venous shunt persists.

In severe pre eclampsia, narrow uterine arteries and minimal arterio-venous shunts are seen.

Here is a broader view of what happens in pre eclampsia

- In normal pregnancy, maternal spiral arteries are invaded by fetal origin cytotrophoblast which transforms them from resistance vessels (small caliber) to capacitance vessels (high caliber).
- This change to capacitance vessels provides a good utero-placental perfusion, for the developing fetus.
- Simultaneously these cytotrophoblasts not only invade but also differentiate from epithelial phenotype to endothelial one, known as “vascular mimicry” or “pseudovasculogenesis”.
- In preeclampsia, vascular remodelling fails to occur, that is, there is noadequate invasion of spiral arteries by cytotrophoblast. This renders spiral arteries in Pre eclampsia to remain as resistance vessels (small caliber)

These vascular changes in normal pregnancy and Pre eclampsia are illustrated in figure 6 and figure 7 respectively.

FIGURE 5:

ANATOMY OF UTERINE AND PLACENTAL VASCULATURE

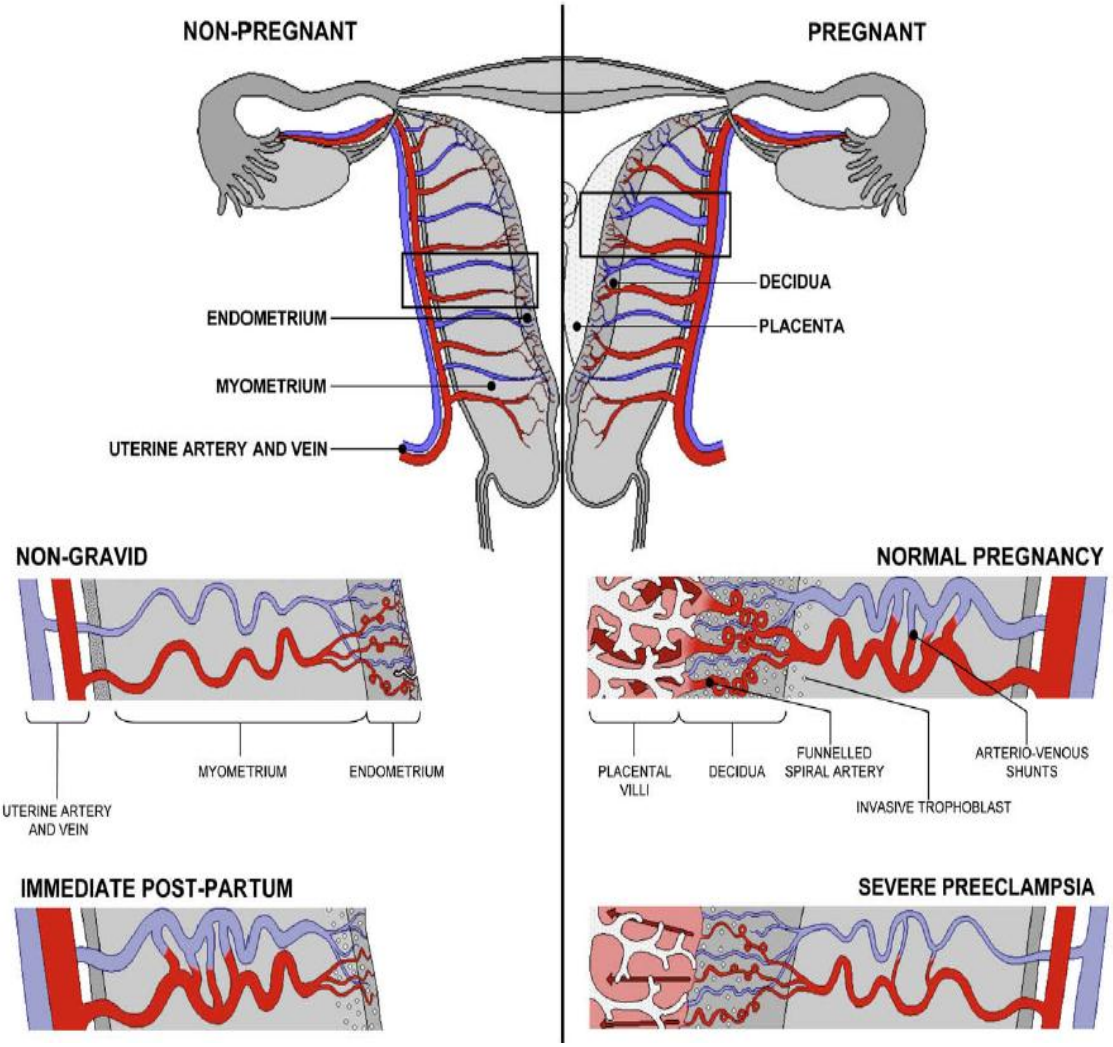


FIGURE 6:

COMPLETE CYTOTROPHOBLAST INVASION IN NORMAL PREGNANCY.

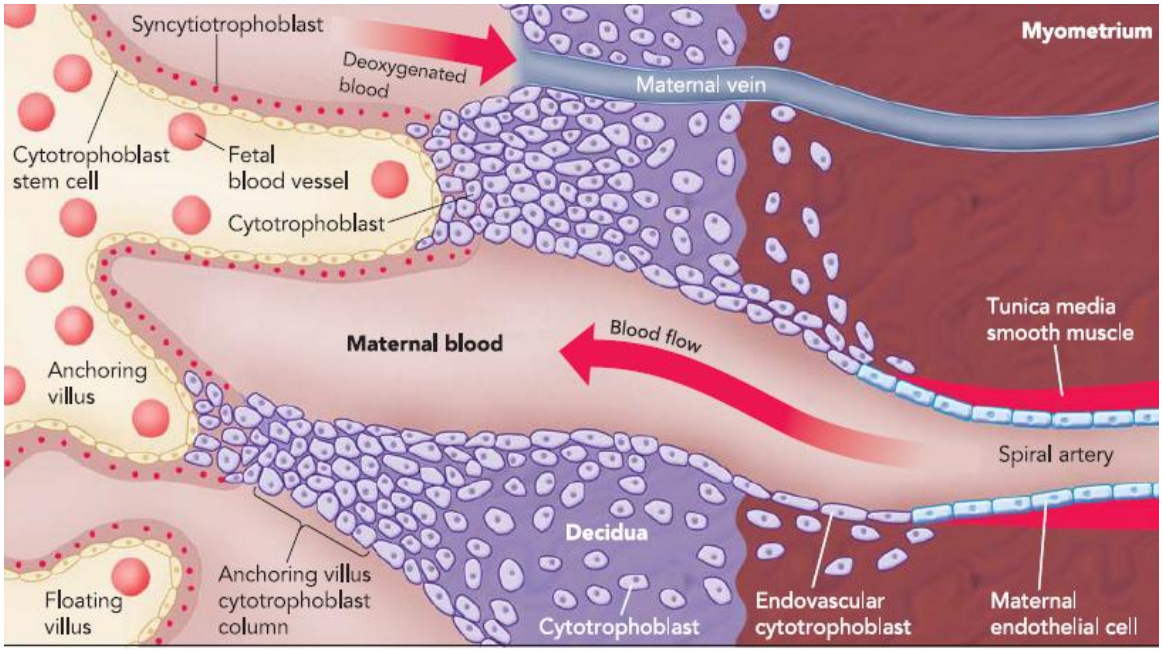
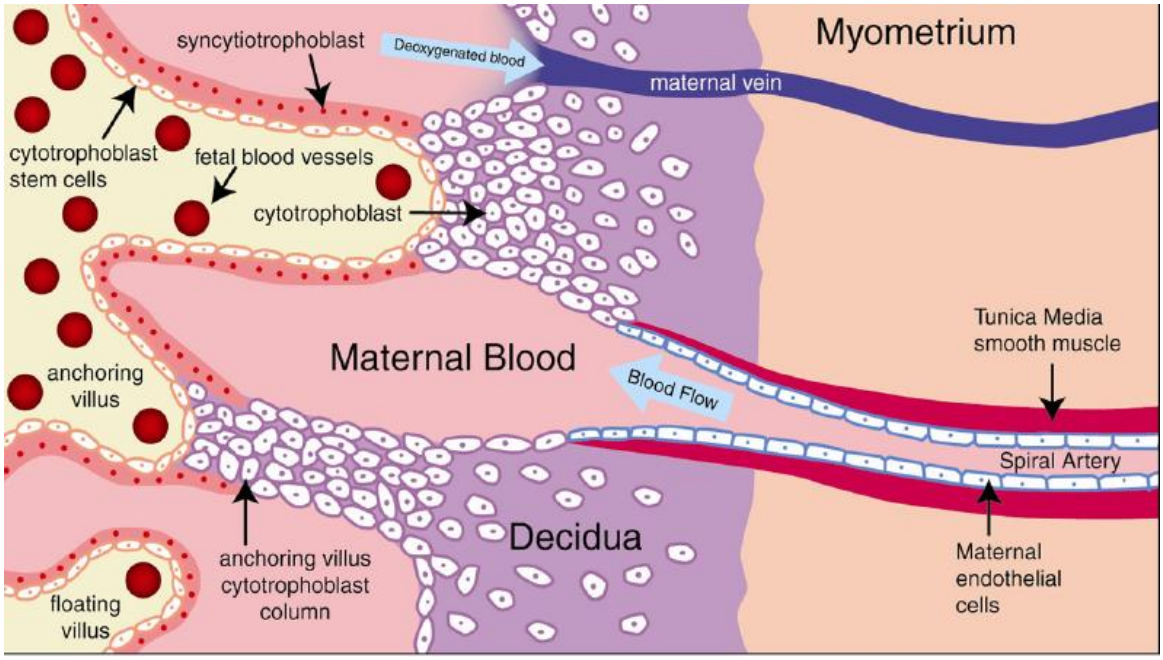


FIGURE 7:

SHALLOW CYTOTROPHOBLAST INVASION IN PRE ECLAMPSIA:[(10)]



III.PLACENTAL ANGIOGENIC FACTORS:

Endothelial dysfunction is caused due to the imbalance between the expression of anti-angiogenic and angiogenic factors.

According to literature, some angiogenic factors are

- Vascular growth factor (VEGF)
- Placental growth factor (PlGF)
- Endoglin
- Angiopoietins
- Transforming growth factor (TGF-β)
- Fibroblast growth factor.

Among these, the first three factors play a key role in pre eclampsia, are discussed here.

VASCULAR GROWTH FACTOR:

VEGF is a dimeric glycoprotein synthesised from cytotrophoblast, T cells, macrophages.[10] Its various isoforms are VEGF-A, VEGF-B, VEGF-C, VEGF-D, PlGF. These act on three different cellular receptors namely Flt-1(VEGFR1), Flk-1(VEGFR), Flt-4(VEGFR3).

VEGF and PlGF are essential for survival of endothelial cells and to maintain maternal vascular homeostasis as well as for embryonic angiogenesis and vasculogenesis.[11]

Apart from this, it has a direct role in systemic vasodilation by stimulating nitric oxide dependent pathway.

The angiogenic property of VEGF, PlGF is inhibited by anti-angiogenic property of sFlt-1.

So what is sFlt-1? And how does it differ from Flt-1?

sFlt-1 is synthesized in syncytiotrophoblast, by mRNA splicing of Flt-1 gene.

Cellular receptor Flt-1 normally contains three domains, namely extracellular, transmembrane and cytoplasmic tyrosine kinase domain.

In sFlt-1, transmembrane and cytoplasmic domains are absent, which makes this receptor lose its ability to bind to VEGF and PlGF within the cell, but it can bind to growth factors in maternal circulation.

In pre eclampsia, there is increased expression of sFlt-1, it antagonises VEGF and PlGF by binding them in maternal circulation and inducing maternal endothelial dysfunction.[11]

To summarize the role of sFlt-1 as an anti-angiogenic factor is, it inhibits the activity of both VEGF and PlGF.

TRANSFORMING GROWTH FACTOR – β (TGFβ)

It is a family of ubiquitous growth factors with diverse functions, one of which is angiogenesis.

The intracellular signalling is initiated by binding of TGFβ with activin like kinase (ALK5) receptor. In vascular signalling, it requires co-receptor Endoglin (ENG), which is expressed in placental syncytiotrophoblast.

Soluble endoglin (sENG) is a splicing variant of ENG, which reduces the binding of TGFβ with ALK-5, anti-angiogenic factors.

IN NORMAL PREGNANCY

Normal circulating levels of TGFβ and VEGF bind to corresponding receptors and enters cytosol.Within cell, physiological levels of TGFβ and VEGF maintains normal vascular homeostasis as shown in figure 8.

IN PRE ECLAMPSIA

Antiangiogenic factors such as sENG and sFLT-1 binds to circulating levels of TGFβ and VEGF thereby decreasing its entry into cytosol. Hence decreased levels of TGFβ and VEGF causes endothelial dysfunction and impaired vascular relaxation in Pre eclampsia as illustrated in figure 9.

FIGURE 8: ANGIOGENESIS AND ANTIANGIOGENESIS FACTORS IN NORMAL PREGNANCY

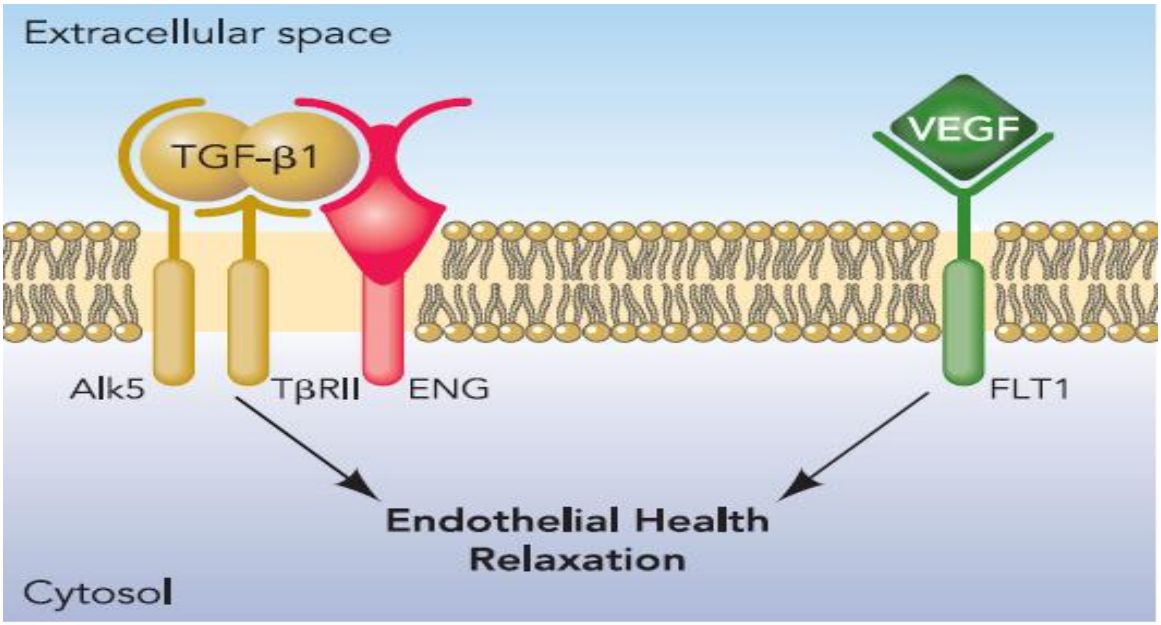
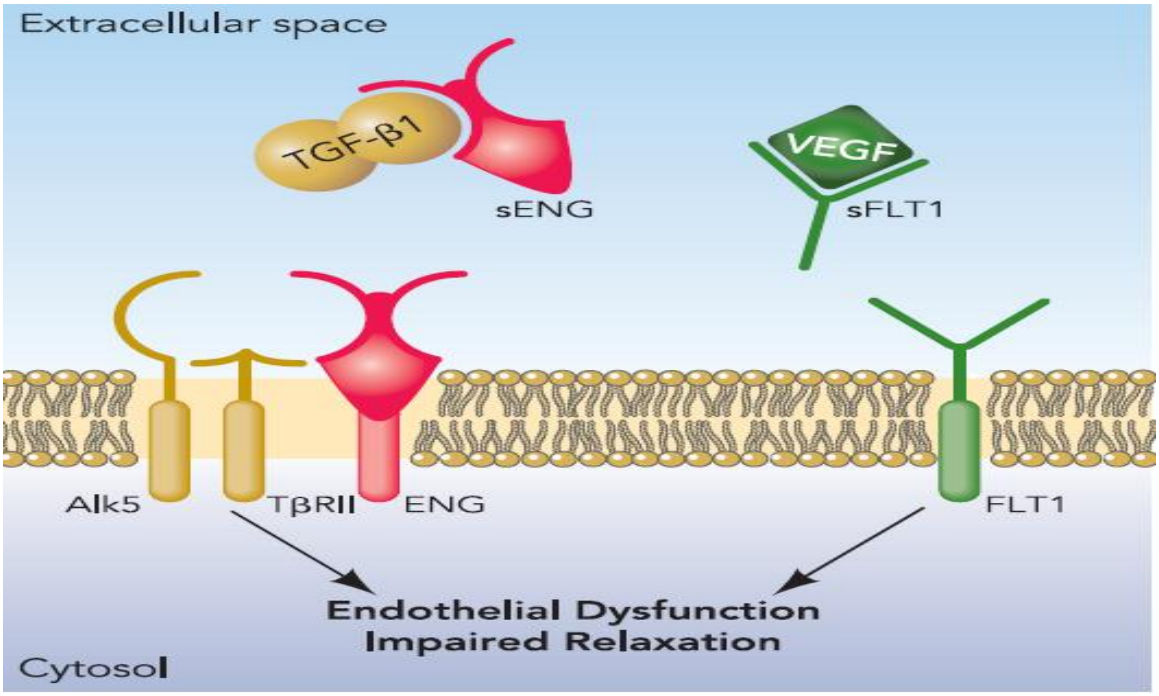


FIGURE 9:

ANGIOGENESIS AND ANTIANGIOGENESIS FACTORS IN PRE ECLAMPSIA.



IV. EXCESSIVE OXIDATIVE STRESS.

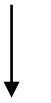
Oxidative stress results due the imbalance between reactive oxygen species and antioxidant defence mechanism. In pre eclampsia, improper placentation causes intermittent hypoxia and reoxygenation resulting in oxidative stress. [13]

One of the earliest insults that occurs in Pre eclampsia is a defective response to oxidative stimulus.

XANTHINE OXIDASE ENZYME:

Reoxygenation and intermittent hypoxia.

Stimulates activation of enzyme Xanthine oxidase (Expressed in syncytiotrophoblast and stromal villous).



Through receptor N-formyl methionyl leucyl phenylalanine.

Ultimately results in free radical induced tissue damage.

NADPH OXIDASE ENZYME:

Increased vascular stress in feto-placental interface, elevated levels of maternal cytokine concentration, increased sensitivity to Angiotensin II.

↓

Acts as a stimulus for NADPH oxidase.

↓

Through receptor phorbol 12 myristate 13-acetate.

↓

Causes tissue damage because of free radicals.

HEME OXYGENASE ENZYME:

It is a heme degradation enzyme in three isoforms.

↓

Out of which HO-1 is expressed more in non-invasive trophoblast.

↓

Normally it increases VEGF/sFlt ratio favouring angiogenesis.

↓

In pre eclampsia, HO-1 is decreased.

Similarly, enzyme activity of superoxide dismutase and glutathione peroxidase are reduced in pre eclampsia. As antioxidants are reduced, it increases reactive oxygen species (ROS), which causes hazardous complications like protein carboxylation, DNA oxidation, lipid peroxidation.

Lipid peroxidation of cell membrane makes it lose its fluidity, whereby protein permeability to membrane is increased, leading to proteinuria and edema.

Excessive oxidative stress is summarized in figure 10.

V. MALADAPTATION OF IMMUNE SYSTEM:

One of the hypothesis, is that Pre eclampsia is an immune rejection response of mother to genetically foreign fetus.[12] Th1 cytokine is pro-inflammatory whereas Th2 is anti-inflammatory.

In normal pregnancy, Th2 polarization occurs which is shift of T-cell phenotype more towards Th2 than Th1. In pre eclampsia, failure of Th2 polarization results in shift towards Th1phenotype causing improper trophoblast invasion as shown in figure 11.

Other theories are, syncytiotrophoblast exosomes and micro vesicles, which are rich in endoglin and sFlt-1 instigates an inflammatory response.[5]

FIGURE 10:

OXIDATIVE STRESS IN PRE ECLAMPSIA

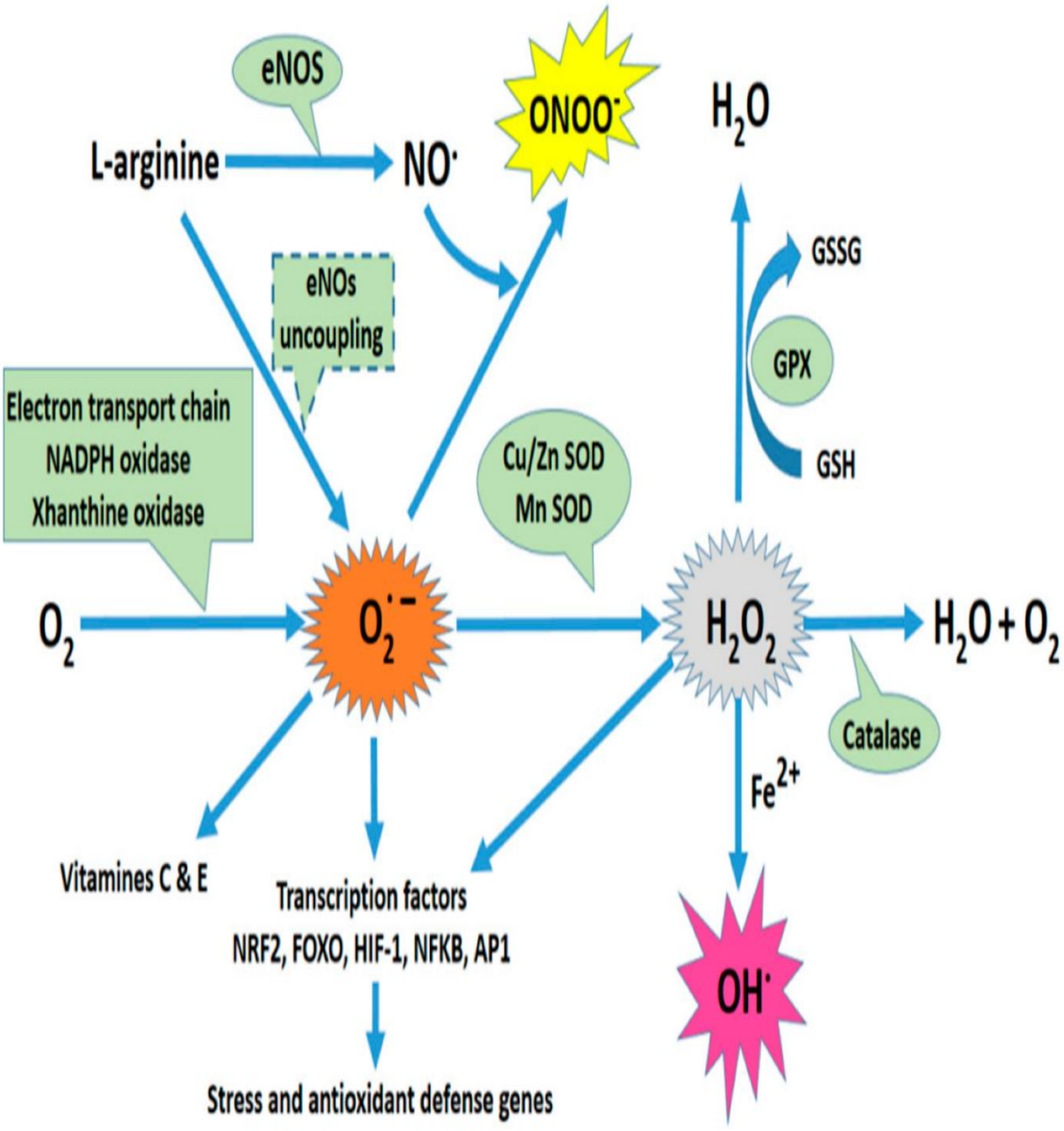
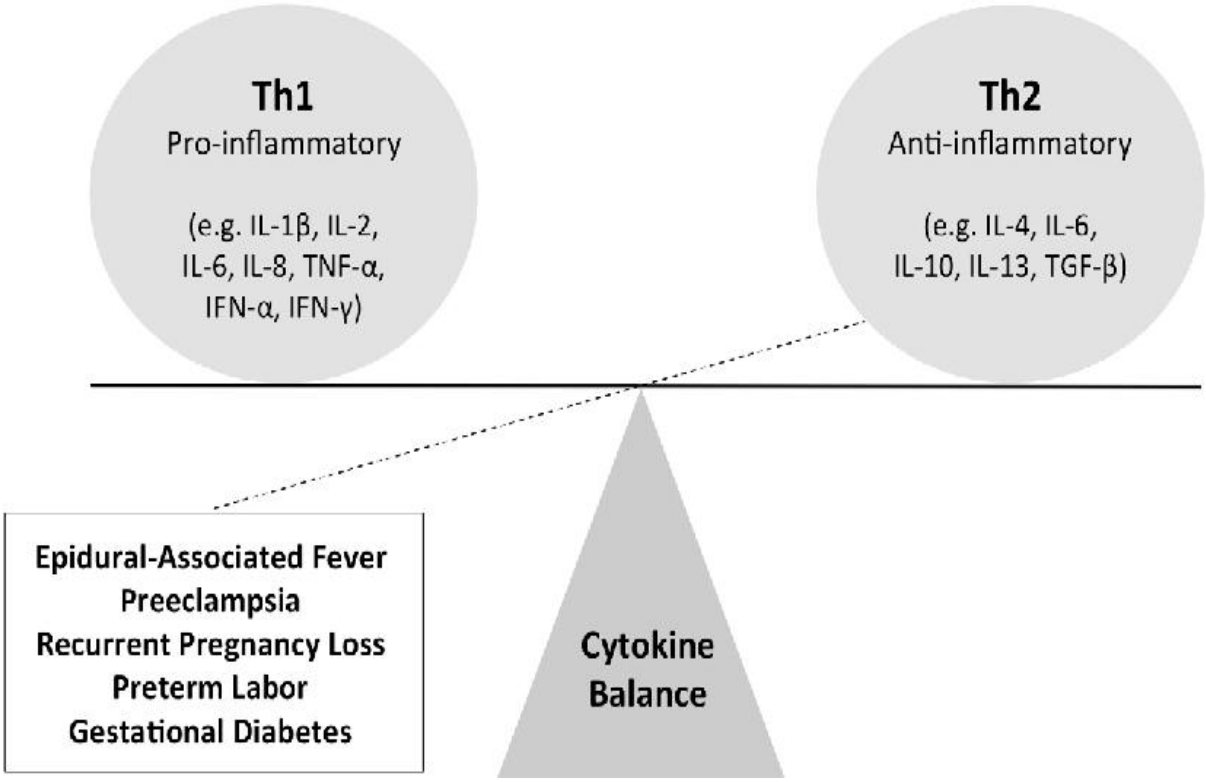


FIGURE 11:

ABSENCE OF Th2 POLARIZATION IN PRE ECLAMPSIA.



These micro vesicles activate mononuclear cells, releasing proinflammatory cytokines. According to literature, IL-10 acts as a mitigator of maternal syndrome as it neutralises proinflammatory cytokines like ET-1, Reactive oxygen species released from placenta, AT1-AA.

uNK cells are down regulated in Pre eclampsia which is necessary for normal trophoblastic invasion and spiral artery remodelling.

The immunological events related to Pre eclampsia are summarised in figure 12.

Placenta secrete two cytokines - IFN γ, Th1

↓

Inhibits trophoblast invasion by arresting production of activin-promoted MMP

↓

DC and Treg lymphocytes regulate cytokine production

↓

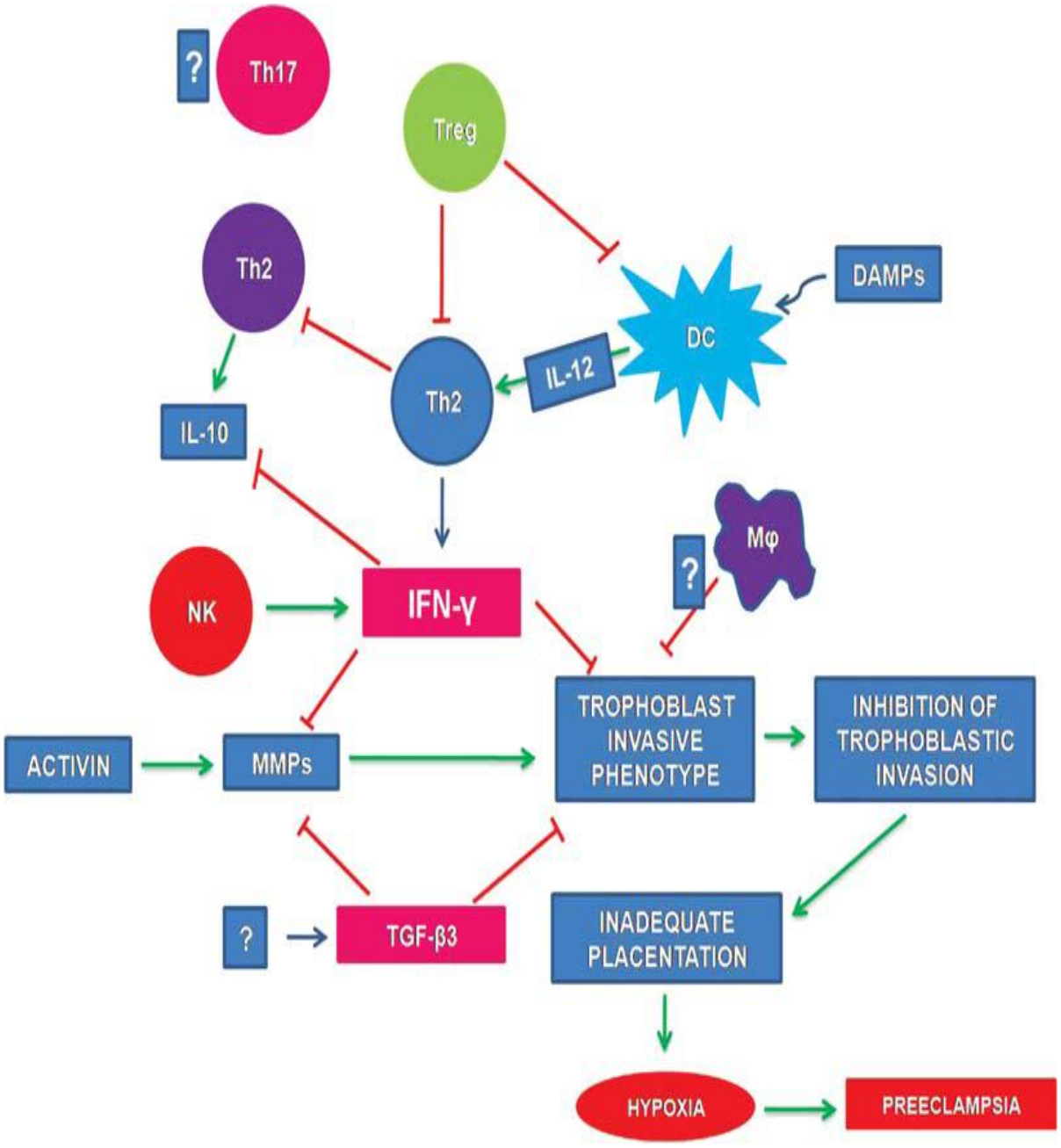
Macrophages (MΦ) also causes poor trophoblastic invasion.

↓

All these cascades of events ultimately result in poor trophoblastic invasion, hypoxia, pre eclampsia.

FIGURE 12:

IMMUNOLOGICAL EVENTS IN PRE ECLAMPSIA.



VI.RENIN ALDOSTERONE ANGIOTENSIN- II

Different RAS components namely Angiotensin, Angiotensin converting enzyme play a major role in spiral artery remodelling and decidualization.[13]

In addition, various studies report shows that angiotensin, prorenin and renin-prorenin receptor(PRR) regulates angiogenesis in placenta through the expression of VEGF.[14]

The exact role of RAS in Pre eclampsia is uncertain though circulatory RAS is decreased; hypertension prevails in this disorder.

Widely accepted hypothesis is based on the role of AT1-AA as depicted in figure 13.

So, what is AT-1 AA?!

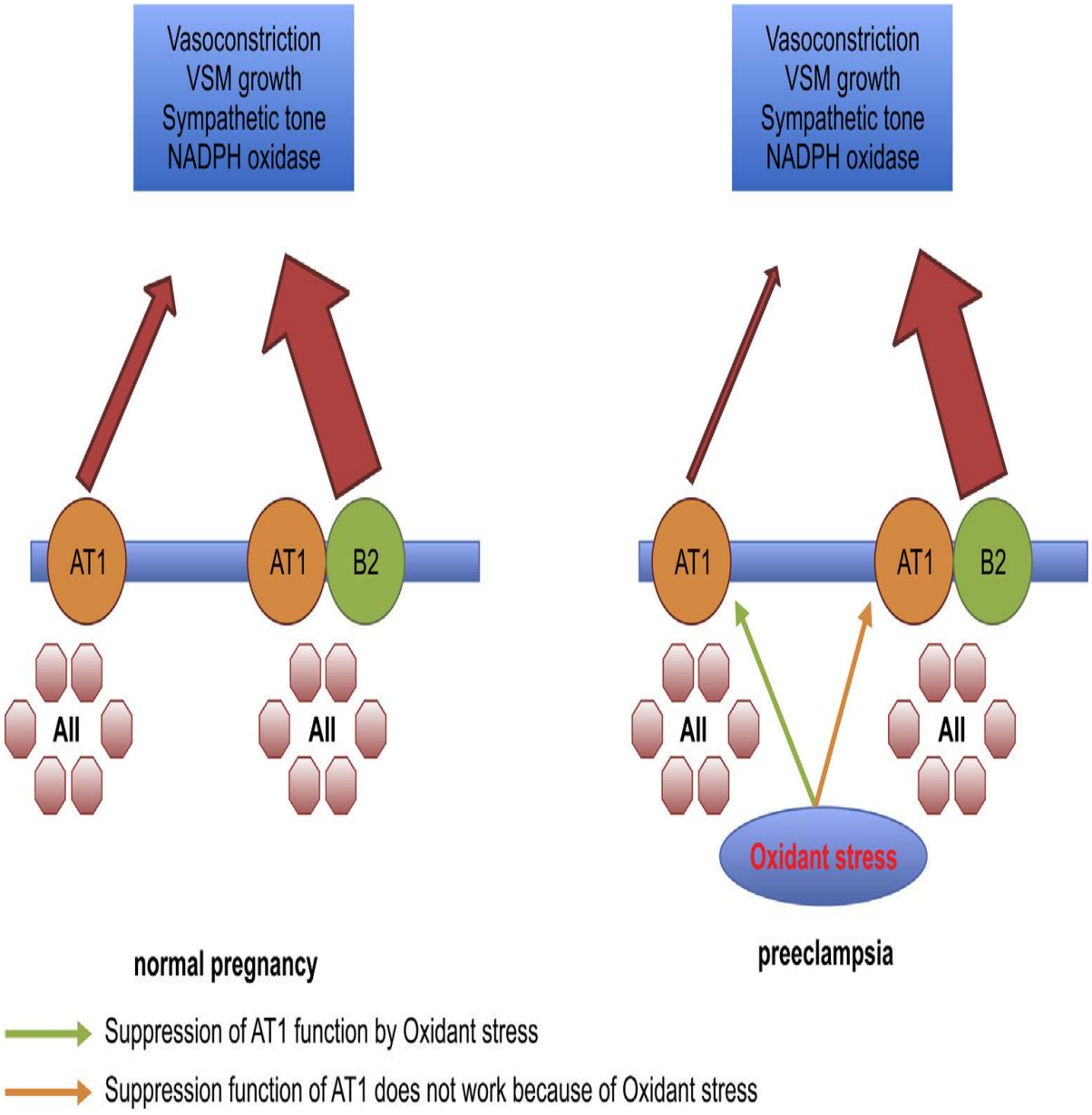
AT-1 AA is Angiotensin II type 1 receptor autoantibody. In 1999 Wallukat et al, first described it in new onset of hypertension in pre eclampsia.

Proposed physiological functions of AT-1 AA are[15]

- It increases the synthesis of sFlt-1.
- Produces Reactive oxygen species by activating NADPH oxidase.

FIGURE 13:

ANGIOTENSIN II RECEPTOR MORPOHOLOGY.



- Increases production of PAI-1 thereby aggravating proteinuria and renal injury.
- Increased ROS produces oxidative stress to placental tissue.
- Increases synthesis of tissue factor, causing accelerated coagulation.
- Increases intracellular calcium.

These various mechanisms substantiate the pathological changes observed in placenta and clinical symptoms. Dechend et al, showed the absence of AT-1 AA in normal conception and non-pregnant hypertensive disorders.

One more interesting hypothesis regarding role of RAA system in pregnancy and Pre eclampsia is explained as follows.

Anton et all, Herse et al, proved that the expression of AT1 and AT2 gene was 5 times more in Pre eclampsia than in normal pregnancy.

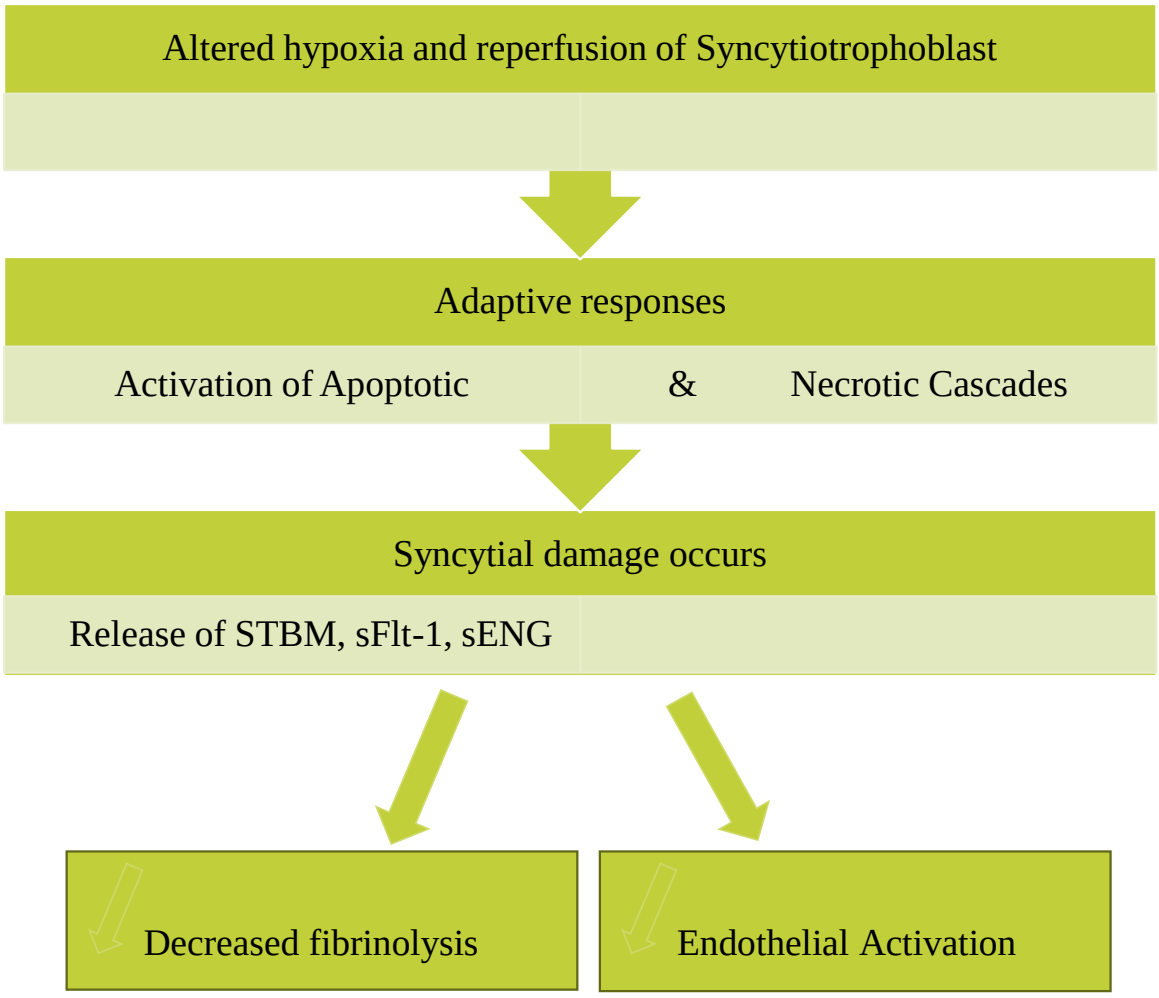
Abdalla et al, conducted a different study focusing on receptors of Angiotensin.

In normal pregnancy, AT1 receptor presents itself as a monomer upon which oxidative stress produces a negative feedback mechanism.

Whereas in pre eclampsia, AT1 combines with bradykinin(B2) to form a heterodimer, thereby becoming resistant to ROS inactivation.[14]

VII. SYNCITIOTROPHOBLAST DEBRIS:

In pre eclampsia, Syncytiotrophoblast microparticles(STBM) are shed in large quantity in maternal circulation. STBM acts as a stimulus for systemic inflamatory response ending up in second stage of Pre eclampsia called Maternal syndrome.



STBM contains placental factors like peroxides, cytokines, annexin V binding microparticles, sFlt-1 and other microparticles.[16] These are increased multifold in pre eclampsia, resulting in oxidative stress, endothelial dysfunction.

Role of STBM in Pre eclampsia are

- To activate blood monocytes and B cells they produce proinflammatory cytokines like IFNγ, TNFα, IL-12, IL-6, IL-8, IL-18.
- And to promote expression of danger molecules responsible for systemic inflammatory response such as HSP-70, HMGB-1, tissue factor. [17]

RECENT UPDATE

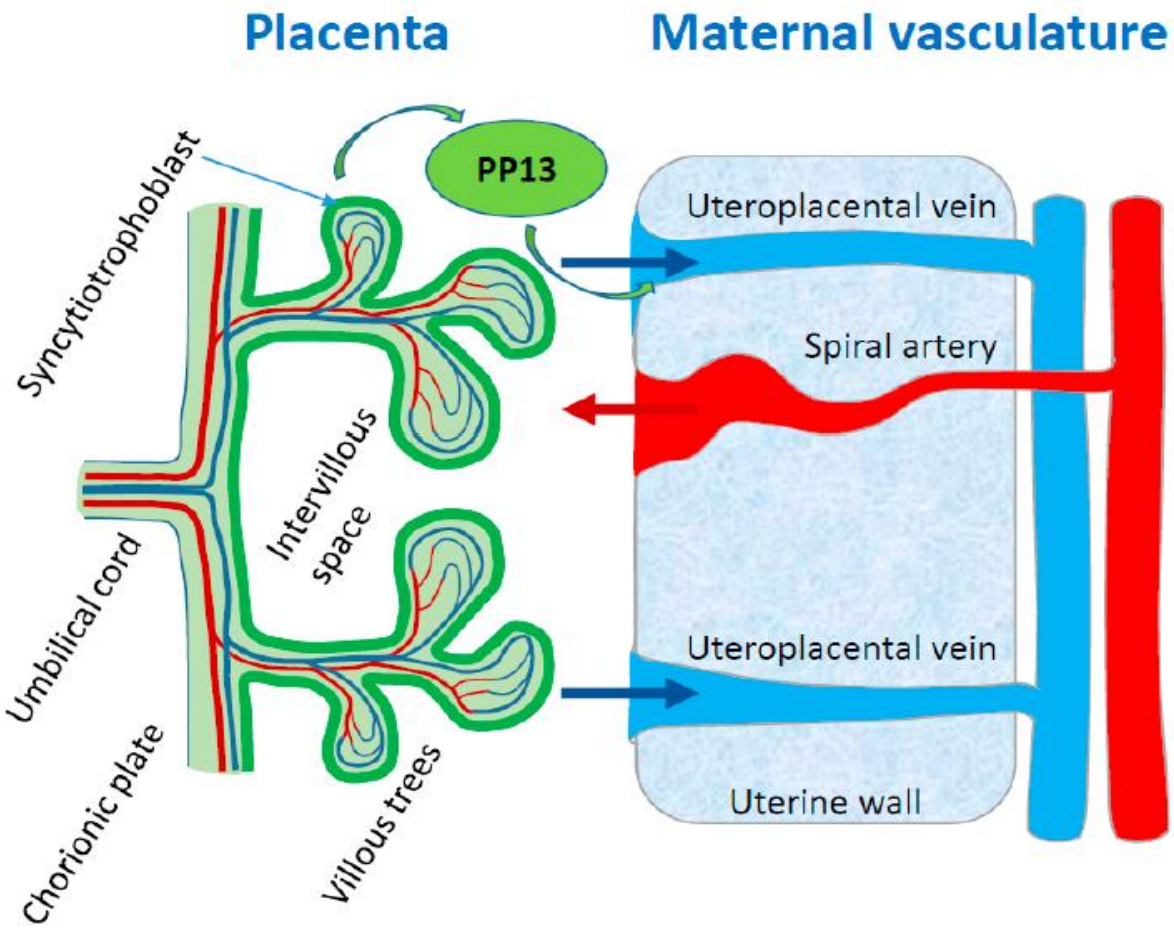
GALECTIN 13(PP13) – ITS ROLE IN PRE ECLAMPSIA- FIGURE 14

Galectin belongs to a class of carbohydrate binding protein, that plays a major role in cell growth proliferation, regulation, differentiation, signal transduction, apopotosis, mRNA splicing etc.

It is released from syncytiotrophoblast and can be detected very early in maternal blood as early as five weeks of gestation.[18]

FIGURE 14:

COMPREHESIVE MODEL EFFECTS OF GALECTIN(PP13) IN MATERNAL VASCULATURE.



The effects of PP13 in maternal vascular system are;

- Vasodilation of uterine vessels.
- Reduction of maternal blood pressure.
- It acts through the Prostaglandin and nitric oxide signalling pathway and increases nutrition and oxygen supply to fetus.

Etiopathogenesis causing pre eclampsia is summarized in the figure 15.

CLINICAL SIGNS AND SYMPTOMS – FIGURE 16

The order of occurrence of signs and symptoms vary from individual to individual.

Most typical symptoms are:

- Headache
- Visual disturbances
- Excessive nausea and vomiting
- Upper abdominal pain
- Decreased urine output

FIGURE 15:

SUMMARY OF ETIOPATHOGENESIS RELATED TO PRE ECLAMPSIA:

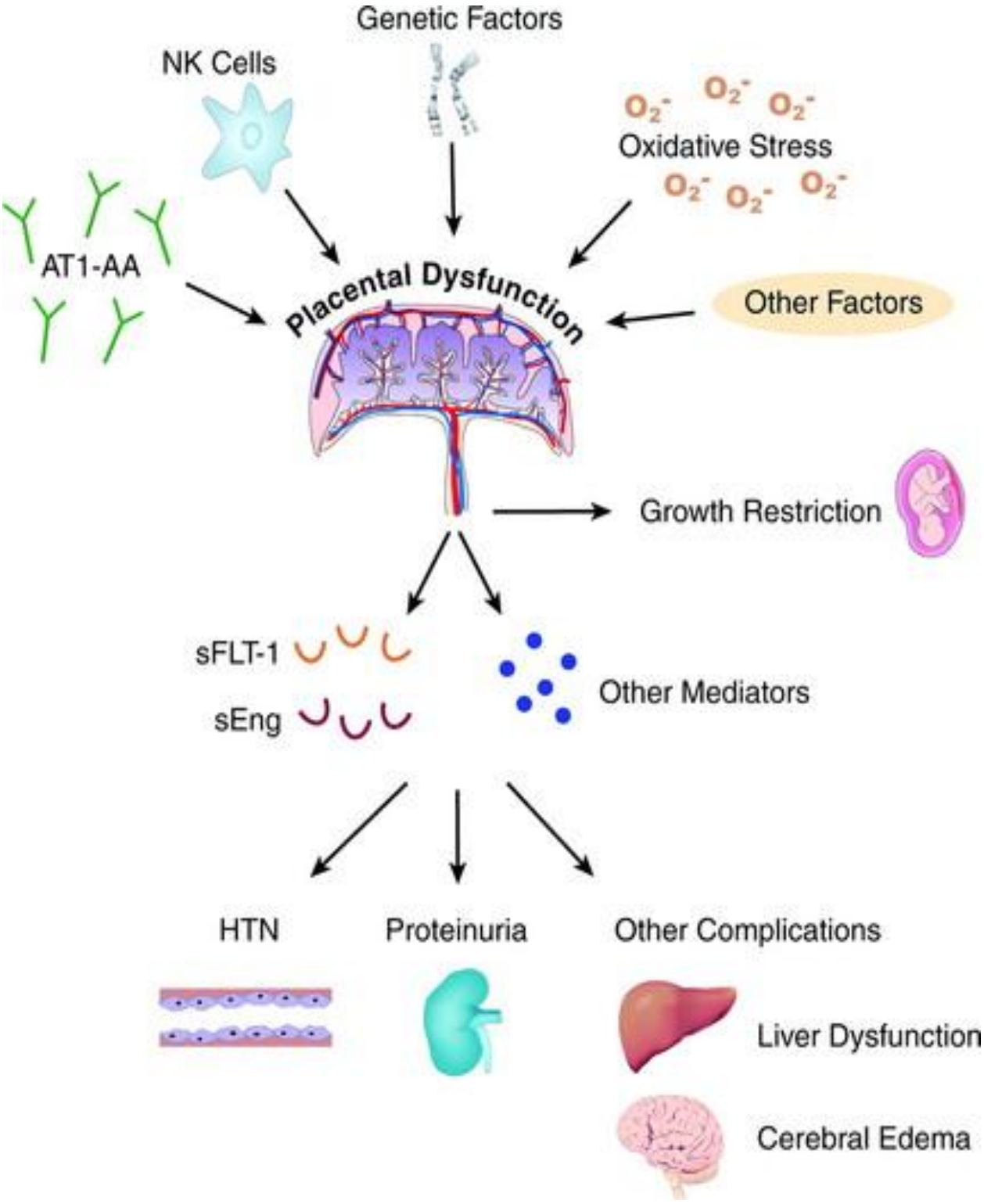
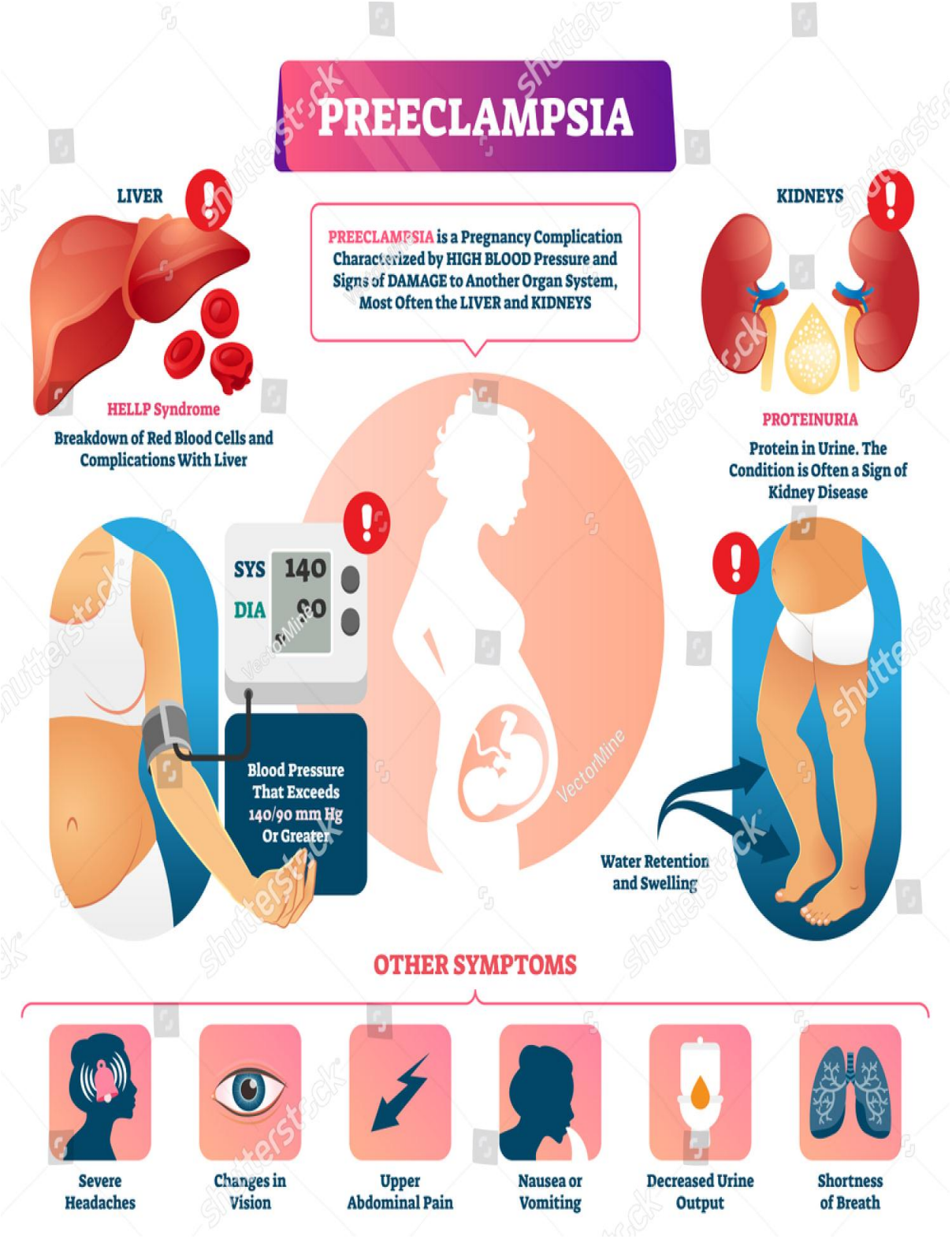


FIGURE 16:

PICTURE ILLUSTRATING SIGNS AND SYMPTOMS IN PRE ECLAMPSIA.



Common Clinical signs elucidated are:

- Uncontrolled hypertension
- Pathological edema in periorbital regions and face
- Rapid gain in weight(> 2kg in a week or > 1kg in 2-3 consecutive weeks)
- Anuria in some cases
- Cerebral symptoms such as hyper-reflexia, generalized convulsions.

Pre eclampsia is labelled as a dreaded disease because it is associated with multiple adverse effects in both the mother and inborn fetus.

TABLE 4:

VARIOUS ADVERSE EFFECTS OF PRE ECLAMPSIA ON MOTHER AND FETUS.

MOTHER	FETUS
Hypertension	Pre term delivery
Cardiovascular disorder	Growth restriction
Seizure	Respiratory distress syndrome
Pulmonary edema	Retinopathy of prematurity
Medical renal disease	Cerebral palsy
Liver failure	Sepsis
Death	Necrotizing enterocolitis

Some untoward complications are

- Eclampsia.
- HELLP Syndrome (Hemolysis, elevated liver enzymes, low platelet count).
- Disseminated intravascular coagulation (DIC) syndrome.
- Posterior reversible encephalopathy (PRES).
- Hemolytic uremic syndrome (HUS).

To prevent these deadly complications, it is necessary for timely interventions such as

- ✓ Ante natal steroids for fetal lung maturity
- ✓ Appropriate anti-hypertensive medication
- ✓ Magnesium sulphate regime for seizure prophylaxis.

To achieve a good maternal and fetal outcome, reliable prediction of Pre eclampsia is necessary. Though clinical examination and reliable international guidelines are available, accurately identifying high risk Pre eclampsia mothers is still an enigma.

To overcome this, a single marker or a combination of multiple markers need to be studied systematically across the world.

BIOMARKERS IN PRE ECLAMPSIA:

Till date, no definite therapy or preventive strategy is present for pre eclampsia. Clinical experience by medical pioneers suggest that early detection, continuous monitoring and appropriate supportive care is beneficial to both the mother and unborn fetus.

As Pre eclampsia is a “Disease of theories” multiple markers are on research such as

- Lactate dehydrogenase
- Free fetal hemoglobin
- Alpha -1- macroglobulin
- Pregnancy-associated Protein A
- Placental Protein 13(Galectin)
- Soluble Endoglin
- Angiogenesis factor (VEGF/PlGF)
- Anti-angiogenesis factor
- Serum Prolactin
- Uric acid
- Proteinuria

My study aims at selecting a good biomarker having good sensitivity and specificity at the same time cost effective.

This study is conducted in Government Coimbatore medical college, Obstetrics and gynecology department. Though various biochemical parameters have been put forward in Pre eclampsia, yet a definitive biomarker is yet to be established.

The following basic biochemical parameters have been performed namely

- Uric acid
- Lactate dehydrogenase
- Alkaline phosphatase
- 24 hours urine protein

These are available in almost all renowned Government medical colleges.

In addition, different parameter done is

- Serum prolactin.

As already mentioned, though multiple parameters are available, a good biomarker is necessary. A good Biomarker needs to fulfill the following characteristics such as:

- Easy to perform
- Repeatable
- Sensitive and specific
- Easily available

Biochemical parameters done in this study are discussed in detail as follows.

PROLACTIN:

Prolactin is a single chain polypeptide hormone, of molecular weight 23KDa secreted in both anterior pituitary (lactotrophs) and extra pituitary sites. It belongs to cytokine family and is made up of 199 amino acids.[19] It is a pleotropic neuroendocrine hormone, secreted in a circadian pulsatile rhythm, reaching a highest peak in early morning.[20]

Since 1933, when Riddle and colleagues named the hormone as Prolactin, it is widely recognised for its role in lactation. Until now, more than 300 actions have been proved across reproduction, immune regulation, metabolic and fluid regulation.[21]

MOLECULAR FORMS OF PROLACTIN – FIGURE 17.

A single gene present in chromosome 6, encodes for the hormone, which constitutes 4 introns and 6 exons.[21]It is made up of 4 antiparallel α helices, and this makes it structurally similar to 2 other hormones namely growth hormone and human placental lactogen.[22]

Major circulating form of Prolactin is monomeric 23KDa, which has good biological activity. Other major forms are big prolactin and macroprolactin which are due to post translational modifications of monomeric mature prolactin. They donot have significant biological activity.

PROLACTIN HORMONE RECEPTOR:

The receptor belongs to hematopoietic cytokine receptor family.

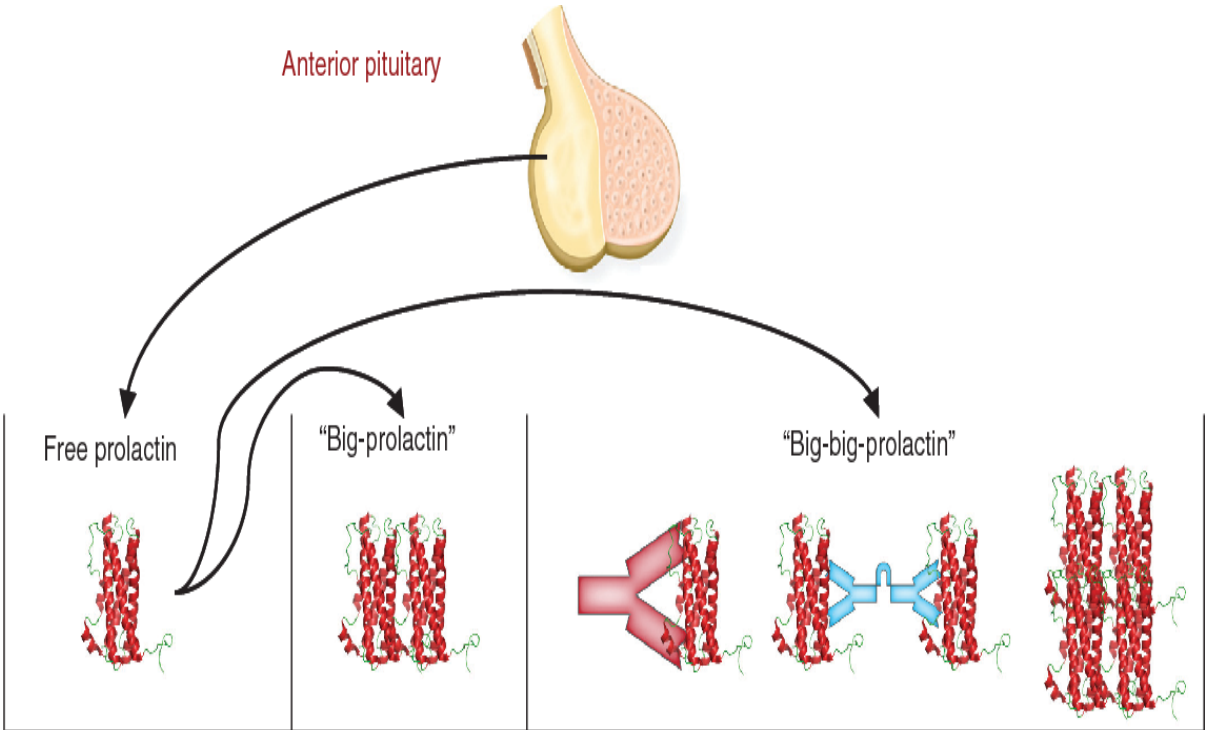
It is made up of 3 domains namely

- Extracellular domain – contains 2 disulfide bridges
- Transmembrane domain
- Cytoplasmic or intracellular signal transducing domain.

PRLR in humans not only binds to prolactin but also to growth hormone and placental lactogen.

FIGURE 17:

ILLUSTRATES STRUCTURE OF MOLECULAR FORMS OF PROLACTIN[22]



Monomeric	Dimeric	IgG-prolactin	IgA-prolactin	Prolactin aggregates
23 kDa	40–60 kDa	>100–150 kDa		
Active	Inactive	Inactive		
60%–90%	15%–30%	0%–8%	0%–2%	0%–3%

MOLECULAR SIGNALLING OF PROLACTIN:

- Prolactin binding to extracellular domain triggers conformational changes which initiates intracellular signal transduction.
- Many kinases are activated like JAK -2, Src family of tyrosine kinase, mitogen activated protein kinase, phosphatidylinositol 3-kinase, PI3-kinase enhancerA and serine/threonine kinase Nek3-Vav2-Rac1 pathway.[23]

REGULATION OF PROLACTIN SECRETION:

Dopamine plays an important role in suppressing prolactin secretion.

Dopamine neurons are located in arcuate nucleus-hypothalamus which are grouped according to anatomical location into

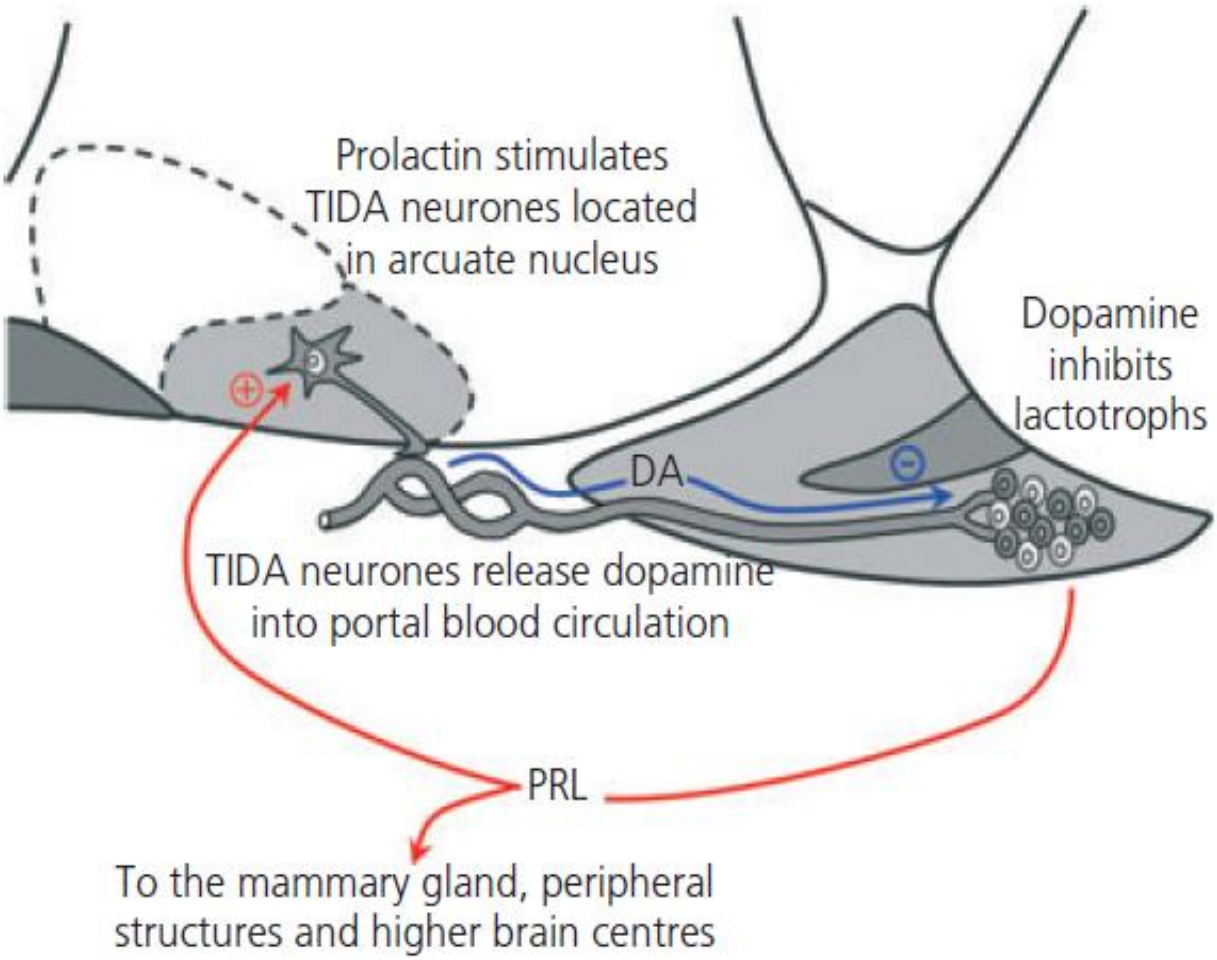
- ✓ Tuberoinfundibular dopaminergic neurons (TIDA)
- ✓ Tuberohypophyseal dopaminergic neurons (THDA)
- ✓ Periventricular hypophyseal dopaminergic neurons (PHDA)

Where major system controlling prolactin, secretion is TIDA.

High Prolactin causes short loop negative feedback by stimulating TIDA which releases dopamine as shown in figure 18. Dopamine acts on anterior pituitary and inhibits lactotrophs and hence prolactin release.

FIGURE 18:

PROLACTIN-SHORT LOOP NEGATIVE FEEDBACK REGULATION :[25]



SIGNAL TRANSDUCTION PATHWAY IN PROLACTIN HORMONE – FIGURE 19.

Among various kinase pathways involved in prolactin, JAK/STAT pathway is described in detail here.

Prolactin binding with receptor induces conformational change

↓

This triggers phosphorylation of JAK2 and recruits cytoplasmic transcription factor STAT5b

↓

Eventually STAT5b gets phosphorylated becomes dimers and are translocated to the nucleus.

↓

In nucleus it binds to regulatory elements and modifies transcription.

↓

There is increased TH mRNA expression and dopamine synthesis.

PROLACTIN IN NORMAL PREGNANCY - FIGURE 20.

In early pregnancy, there is auto regulation of prolactin by short loop negative feedback. But as gestational week progresses, towards late pregnancy, inhibitor TIDA neurons become unresponsive to prolactin leading to high values.

FIGURE 19: PROLACTIN – SIGNAL TRANSDUCTION PATHWAY.[26]

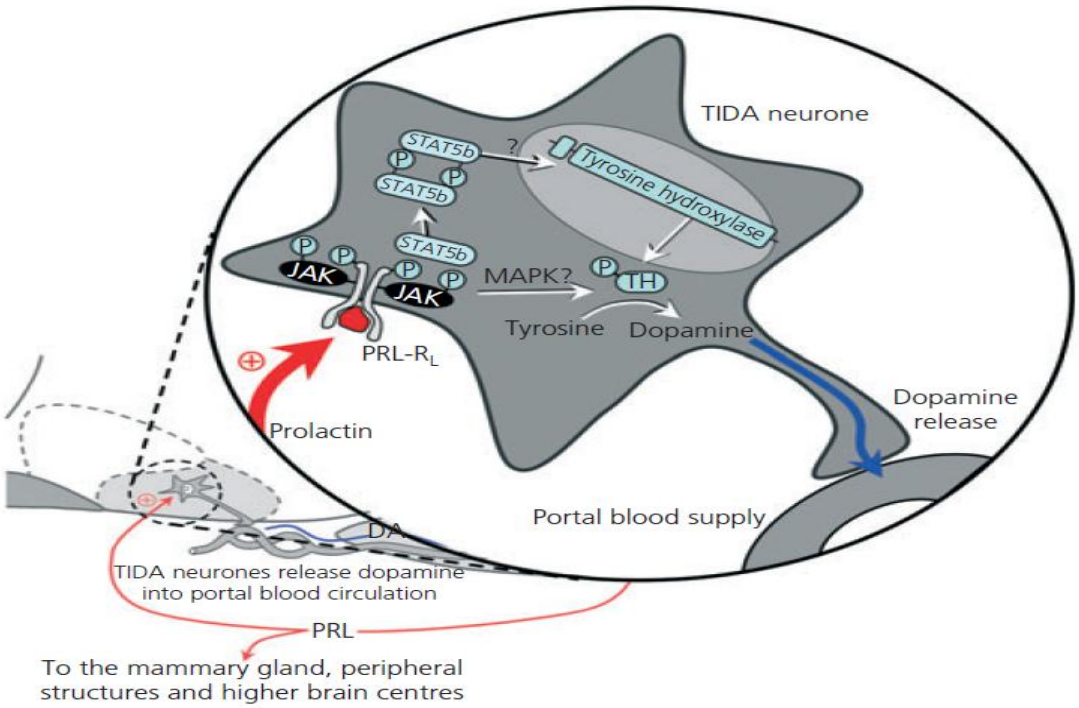


FIGURE 20: INCREASED PROLACTIN IN LATE PREGNANCY.

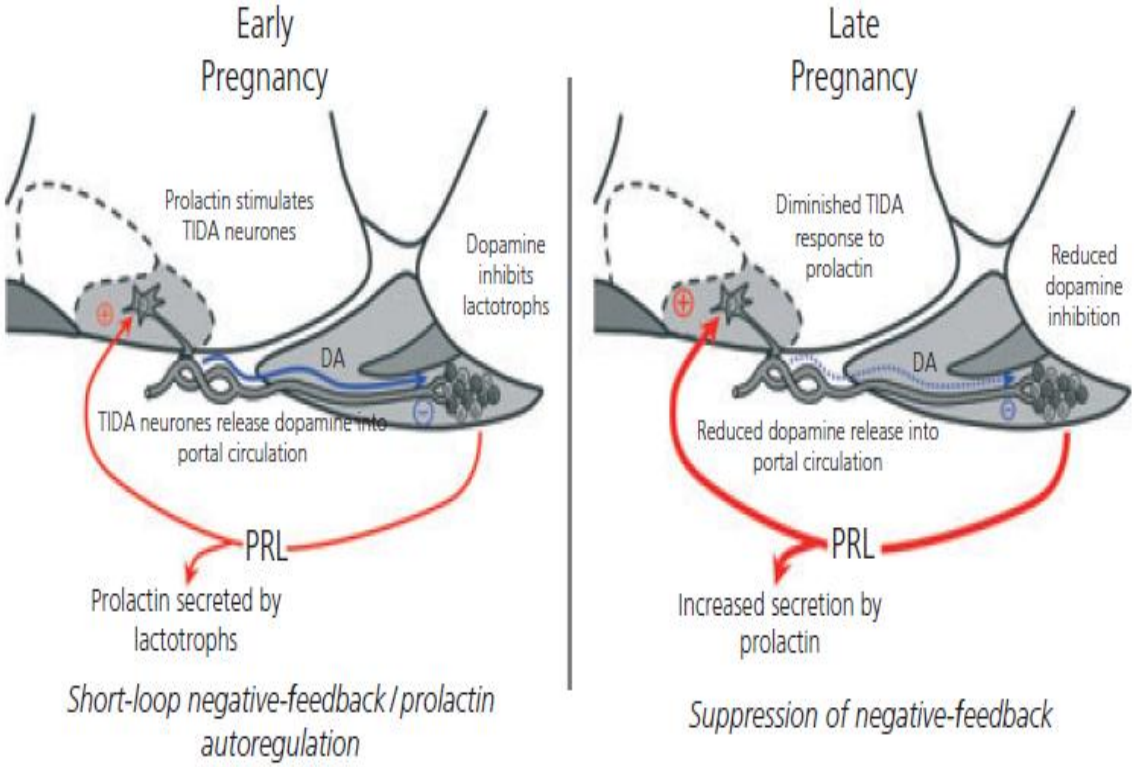


TABLE 5:

SERUM PROLACTIN NORMAL RANGE

CONDITION	S.I UNIT ng/ml
Non-pregnant female	0-19
1st trimester	30-210
2nd trimester	110-330
3rd trimester	140-370

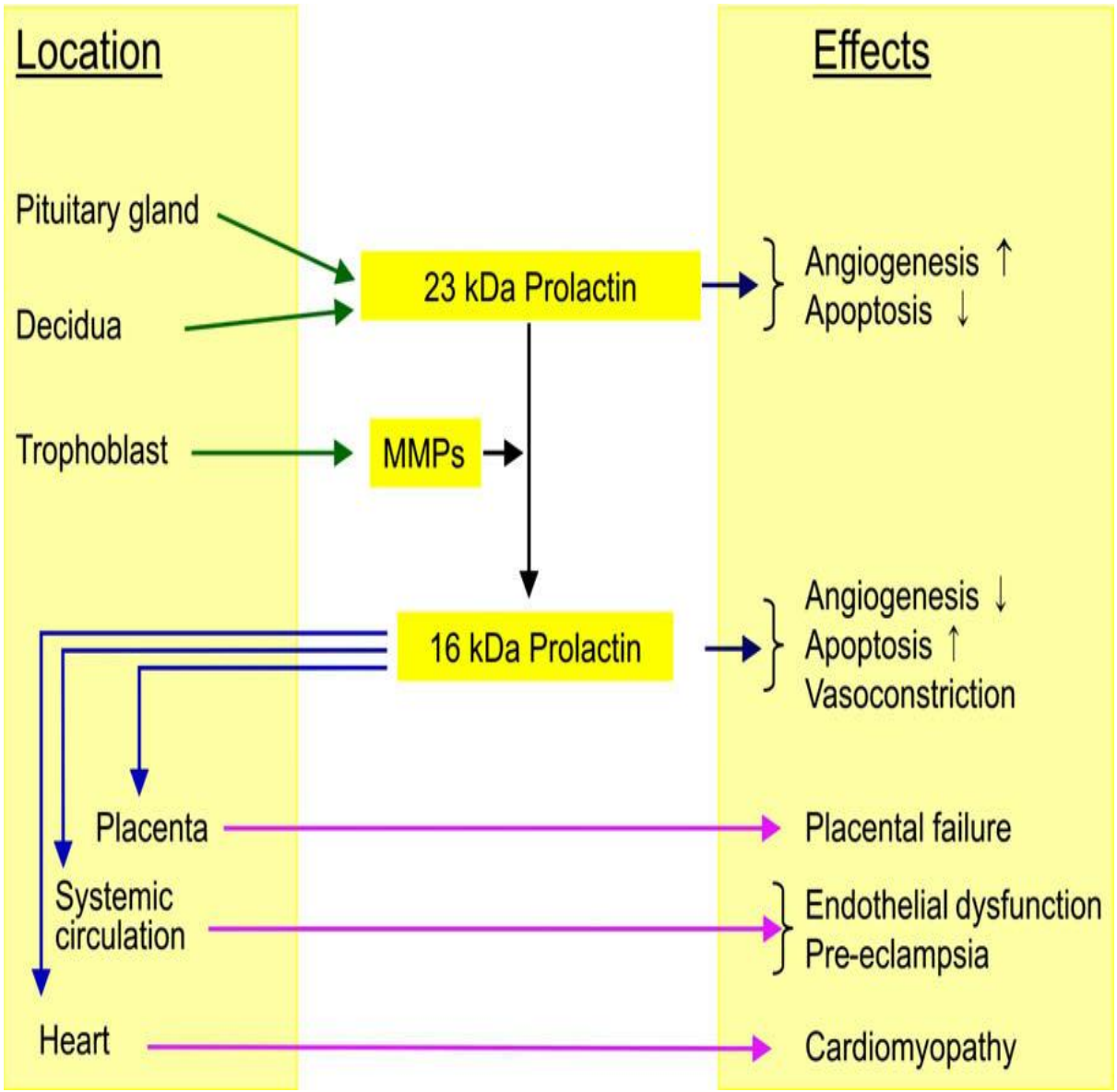
NORMAL BIOLOGICAL FUNCTIONS OF PROLACTIN:

- In lactation- milk synthesis and maintains lactation
- In reproduction
- In immune system- cytokine like immune response
- Osmoregulation -increases salt and water absorption

PROLACTIN IN PRE ECLAMPSIA:

It is prolactin fragments responsible for antiangiogenic and anti-vasodilatation effects as shown in figure 21.[24] Prolactin undergoes proteolytic cleavage by enzymes such as cathepsin D, matrix metalloproteinases and bone morphogenetic protein-1.[25–27]

FIGURE 21: ROLE OF PROLACTIN FRAGMENTS IN PRE ECLAMPSIA[21]



In pre eclampsia, cathepsin D is upregulated, which cleaves mature prolactin (23KDa) into its fragments namely 14 KDa and 16 KDa.[28]

This N-terminal prolactin fragment 16 KDa has potent antiangiogenic property by blocking VEGF and PlGF.

It plays a major role in Pre eclampsia complications, such as postpartum cardiomyopathy.

Antiangiogenic derivative of prolactin, is not confined to 16KDa fragment alone, but also includes Vasoinhibins – a novel family of hormones.[29,30]

An increased, dysregulated vasoinhibin synthesis from placenta is linked to etiology of pre eclampsia, gestational diabetes and various fetal growth abnormalities.[29]

ROLE OF URIC ACID IN PRE ECLAMPSIA:

- Uric acid decreases nitric oxide synthesis from endothelial cells, which is necessary for good trophoblastic invasion and implantation.[31]
- Hyperuricemia is attributed to decreased renal clearance, and hypertension is due to increased renin activity.[32]
- It directly causes endothelial dysfunction by activating certain proinflammatory markers.[33]

LACTATE DEYDROGENASE IN PRE ECLAMPSIA:

- LDH is an intracellular enzyme, in pre eclampsia, increased LDH correlates with cellular death.
- Moderately elevated LDH (600-800 IU/l) is associated with abruptio placenta, cerebrovascular accident.[34]
- Severely elevated LDH (>800 IU/l) are linked with HELLP syndrome, pulmonary embolism, renal failure, metabolic encephalopathy.[35]

ALKALINE PHOSPHATASE:

Human placental trophoblast secrets Heat stable alkaline Phosphatase (HSAP).

This abnormal value of heat stable alkaline precedes at least 2-3 weeks ahead of pre eclampsia, and estimating this parameter can help in preventing impending fetal complications.

PROTENURIA:

- In pre eclampsia, glomerular endothelial leakage causes abnormal proteinuria.[36]
- In recent years, proteinuria has become a symptom of multi-organ involvement rather than a diagnostic criterion.[37]
- Criteria for proteinuria is either $\geq$ 300mg/24-hour urine sample, Or protein creatinine ratio $\geq$ 0.3

Materials and Methods

MATERIALS AND METHODS

SAMPLE SIZE: 100

Case Selection:

CASES: 50 cases of newly diagnosed pre eclampsia.

CONTROLS: 50 cases of Healthy pregnant mother .

INCLUSION CRITERIA:

- Pre eclampsia ante natal mother within 20 to 30 years of age.

EXCLUSION CRITERIA:

- Diabetes mellitus
- Essential hypertension
- Renal disease
- Hypothyroidism
- Multiple pregnancy

STUDY DESIGN: Case control study.

DURATION OF STUDY: One year. (MARCH 2018-MARCH 2019)

SAMPLE COLLECTION:

5ml of venous blood is drawn in a plain vacutainer tube under sterile conditions after fulfilling the selection criteria. Serum is separated by centrifugation and quickly frozen at -20^{0}C and stored until processed**.**

INVESTIGATIONS:

1. Serum prolactin- ELISA technique
2. Serum uric acid
3. Serum alkaline phosphatase
4. Liver function test
5. Renal function test
6. Thyroid profile
7. 24 hours urine Protein
8. Platelet count

ESTIMATION OF SERUM PROLACTIN

BY ENZYME LINKED IMMUNOSORBANT ASSAY:

PRINCIPLE

$$Ag_{(Prl)} + {}^{Btn}Ab_{(m)} \underset{K_{-a}}{\overset{K_a}{\rightleftharpoons}} Ag_{(Prl)} - {}^{Btn}Ab_{(m)}$$

${}^{Btn}Ab_{(m)}$ = Biotinylated Monoclonal Antibody.

$Ag_{(Prl)}$ = Native Antigen.

$Ag_{(Prl)} - {}^{Btn}Ab_{(m)}$ = Antigen- Antibody complex.

K_a = Rate constant of Association.

K_{-a} = Rate constant of Dissociation.

Simultaneously the complex is deposited to the well via high affinity reactions of streptavidin and Biotinylated antibody.

The reaction is as follows

$Ag_{(Prl)} - {}^{Btn}Ab_{(m)} + Streptavidin_{CW} \Longrightarrow$ Immobilized complex(IC).

$Streptavidin_{CW}$ = Streptavidin immobilized on well.

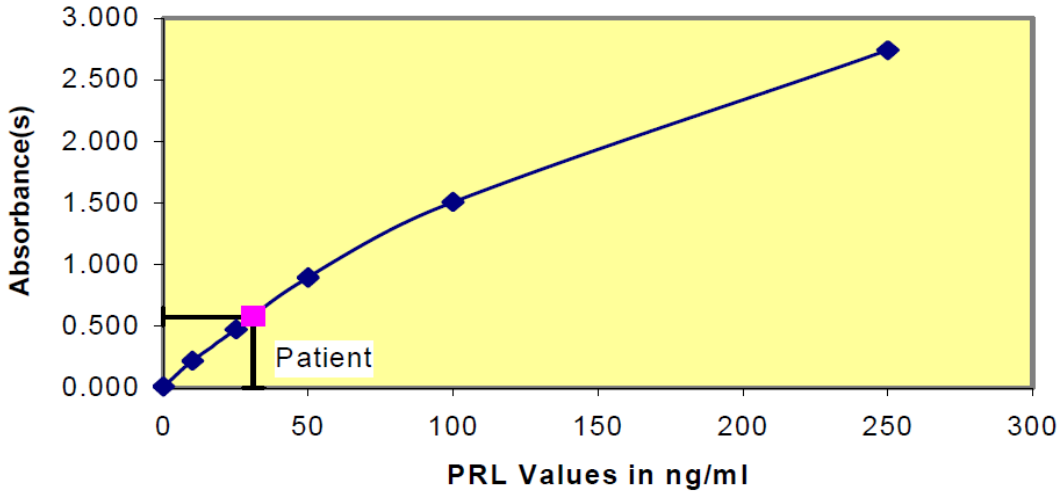
Immobilized complex(IC) = Ag- Ab bound to well.

After incubation period, decantation removes unbound antigen from antibody-antigen complex.

Addition of suitable substrate produces color.

The color produced is directly proportional to the naive antigen.

Several different serum references of known antigen concentration are utilized and a dose response curve is generated.



From the curve, unknown antigen concentration can be ascertained

$$IC + {}^{Enz}Ab_{(x\text{-}Prl)} \underset{K_{-b}}{\overset{K_b}{\rightleftharpoons}} {}^{Enz}Ab_{(x\text{-}Prl)} - IC$$

${}^{Enz}Ab_{(x\text{-}Prl)}$ = Enzyme labelled Antibody.

${}^{Enz}Ab_{(x\text{-}Prl)}$ - IC = Antigen-Antibody Complex.

K_b = Rate constant of Association.

K_{-b} = Rate constant of Disassociation.

PROCEDURE

Format readymade Streptavidin coated microplate well

↓

Add 25 µl of serum sample into assigned well.

↓

Add 100 µl Prolactin Biotin reagent into all wells.

↓

Gently swirl the microplate for 20-30 seconds.

↓

Cover and incubate at room temperature for 30 minutes.

↓

Discard the contents by decantation.

↓

Add 300 µl of wash buffer – wash three times.

↓

Decant the wash

↓

Incubate at room temperature for 30 minutes.

↓

Decant the wash.

↓

Add 350 µl of wash buffer and wash three times.

↓

Decant the wash.

↓

Incubate at room temperature for 15 minutes.

↓

Add 50 µl of stop solution to each well and mix gently for 15-20 seconds.

↓

Read the absorbance within 30 minutes of adding stop solution.

↓

Absorbance was read using Primary wavelength - 450 nm.

Secondary wavelength – 630nm.

ESTIMATION OF GLUCOSE:

Method- Glucose oxidase Peroxidase.

α-D Glucose $\xrightarrow{\text{Mutarotase}}$ β-D Glucose

β-D Glucose + H_20 + 0_2 $\xrightarrow{\text{Glucose oxidase}}$ Gluconic acid + H_20_2

H_20_2 + Phenol +4 aminoantipyrine $\xrightarrow{\text{Peroxidase}}$ Quinonemine complex + $2H_2O$

ESTIMATION OF URIC ACID:

Method - Uricase – POD

$$\text{Uric acid} + O_2 + H_2O \xrightarrow{\text{Uricase}} \text{Allantoin} + CO_2 + H_2O_2$$

$$\text{TOOS} + 4\ \text{Aminoantipyrine} + 2H_2O_2 \xrightarrow{\text{Peroxidase}} \text{Quinonemine complex} + 4H_2O$$

ESTIMATION OF LACTATE DEHYDROGENASE:

Method- Henry et al.

$$\text{Pyruvate} + \text{NADH} \xrightarrow{\text{LDH}} \text{Lactate} + NAD^+$$

DATA MANAGEMENT AND STATISTICAL ANALYSIS:

All data were analyzed using the statistical package for social science (SPSS) 10.0 for Windows program on the computer. All data were given as mean ± standard deviation (SD). The statistical significance was accepted as *p* value <0.05.

Statistical Analysis:

Data entry was made in the Microsoft Excel software in codes and analysis was done with SPSS-20 computer package. Categorical variables are expressed as percentages whereas continuous variables are expressed as mean ± standard deviation. Association between categorical variable was found by **chi-square test** and relationship between continuous variable was assessed by **Student's *t*-test**. P value <0.05 was considered as statistically significant.

Results

RESULTS

TABLE 6

Group	Frequency	Percentage
Cases	50	50.0
Controls	50	50.0
Total	**100**	**100.0**

FIGURE 22

This figure shows equal distribution of study participants as cases and control.

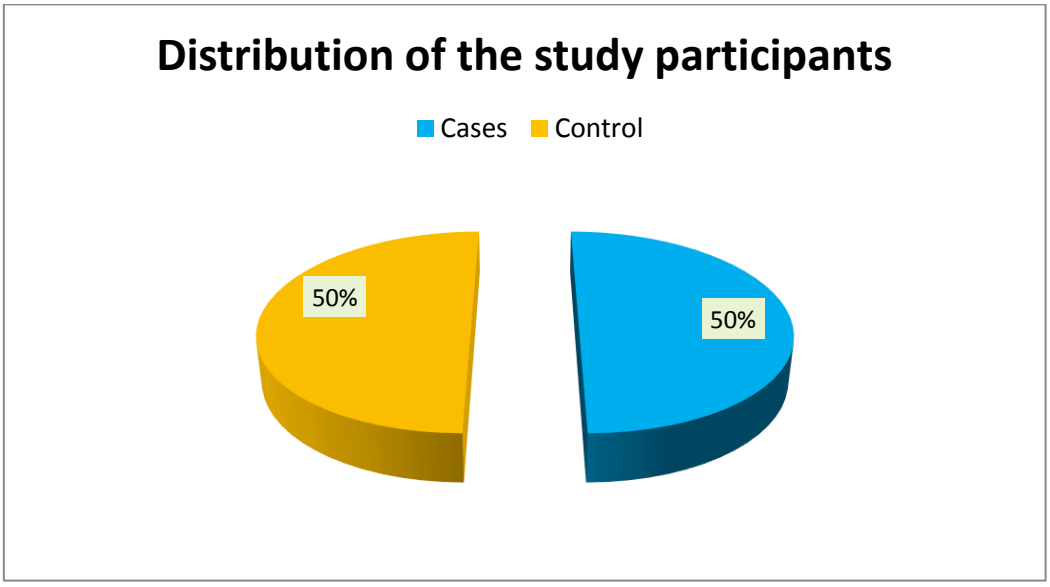


TABLE 7

Age of the study participants	Minimum	Maximum	Mean	Std. Deviation	P value
Cases	17	32	24.50	4.04	0.919
Controls	19	32	24.58	3.78	

The age match between the cases and control is insignificant as p value is 0.919, that is >0.05.

FIGURE 23

MEAN AGE OF THE STUDY PARTICIPANTS IN THE TWO GROUPS

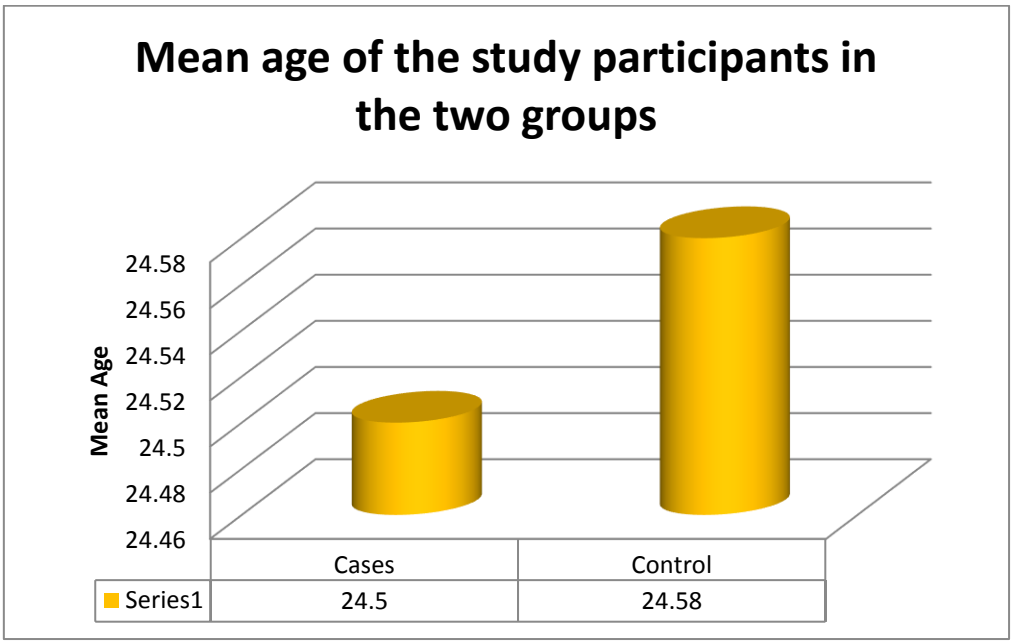


TABLE 8

Age Category	Cases		Controls	
	Frequency	Percentage	Frequency	Percentage
15-20 years	10	20.0	10	20.0
21-25 years	18	36.0	18	36.0
26-30 years	18	36.0	19	38.0
31-35 years	4	8.0	3	6.0
Total	50	100.0	50	100.0

The age category distribution among cases and controls.

FIGURE 24

AGE CATEGORY OF THE STUDY PARTICIPANTS

CASES VS CONTROLS

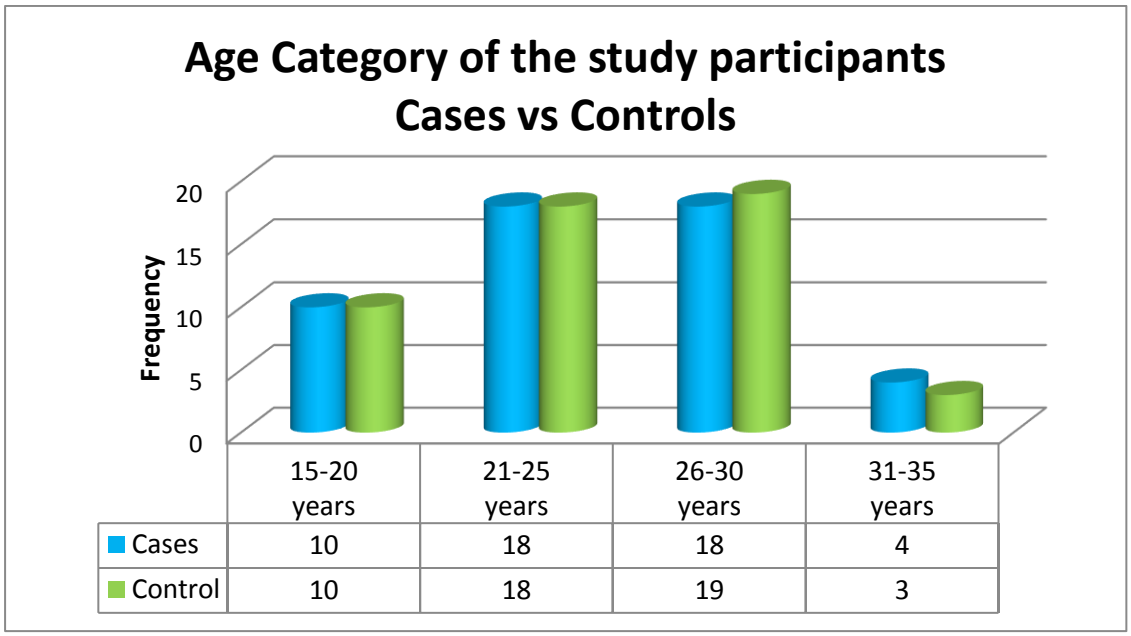


TABLE 9

Liver Function Test	Group	Normal		Abnormal		P value
		N	%	N	%	
Total Bilirubin	Cases	44	88.0	6	12.0	**0.295**
	Control	47	94.0	3	6.0	
Direct Bilirubin	Cases	27	54.0	23	46.0	**<0.001**
	Control	46	92.0	4	8.0	
AST	Cases	7	14.0	43	86.0	**<0.001**
	Control	45	90.0	5	10.0	
ALT	Cases	16	32.0	34	68.0	**<0.001**
	Control	46	92.0	4	8.0	
Alkaline Phosphatase	Cases	6	12.0	44	88.0	**<0.001**
	Control	49	98.0	1	2.0	
Total Protein	Cases	40	80.0	10	20.0	1
	Control	40	80.0	10	20.0	
Sr. Albumin	Cases	35	70.0	15	30.0	**<0.001**
	Control	17	34.0	33	66.0	
Sr.Globulin	Cases	46	92.0	4	8.0	<0.137
	Control	41	82.0	9	18.0	

This table shows, liver function test values and there is significant difference between cases and controls in following parameters namely direct bilirubin, Aspartate aminotransferase, Alanine aminotransferase Alkaline phosphatase and Albumin as the p value is less than 0.05.

Whereas the parameters total bilirubin, total protein and globulin show no significant difference between cases and controls as the p value is more than 0.05.

FIGURE 25

LIVER FUNCTION TEST - CASES GROUP

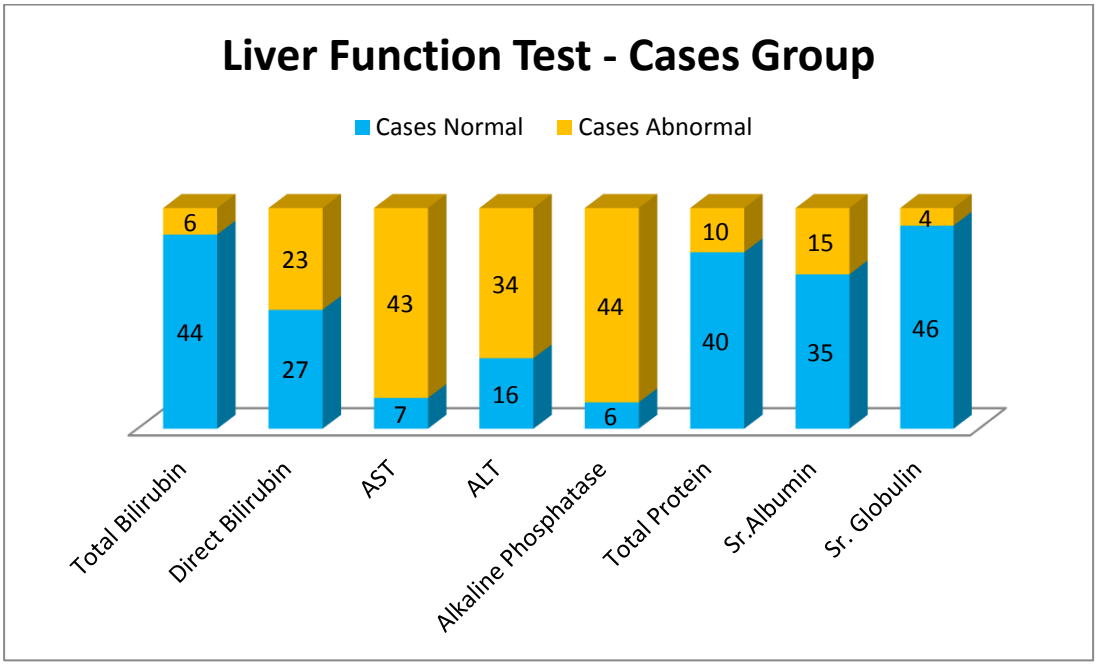


FIGURE 26

LIVER FUNCTION TEST - CONTROL GROUP

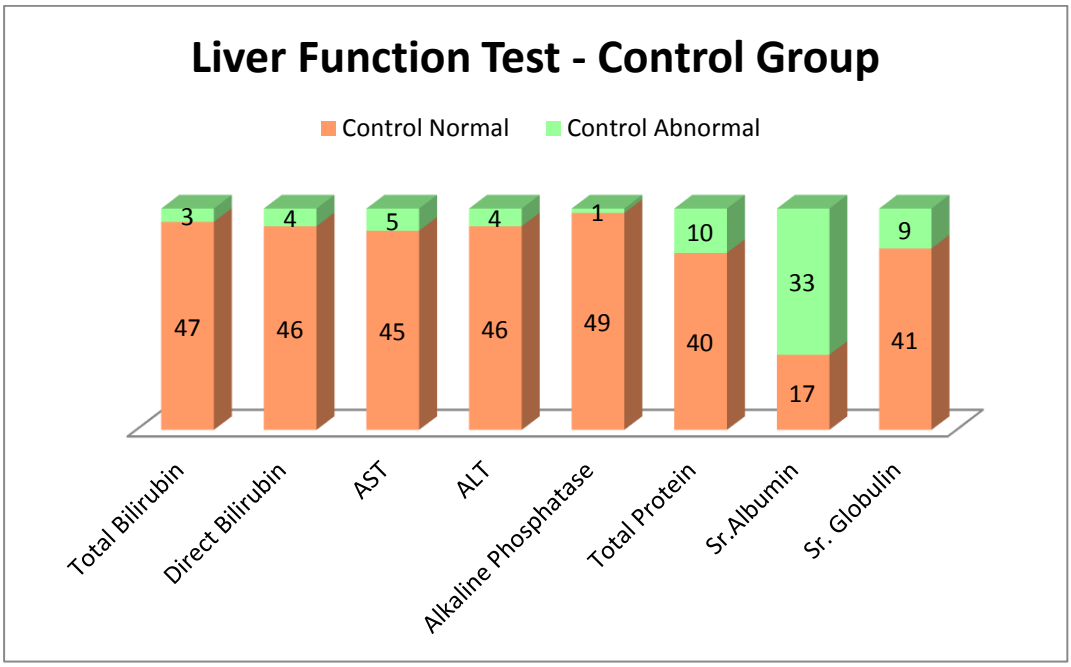


TABLE 10

Variable	Group	Normal		Abnormal		P value
		N	%	N	%	
Prolactin	Cases	3	6.0	47	94.0	**<0.001**
	Control	50	100.0	0	-	
Lactate dehydrogenase	Cases	26	52.0	24	48.0	**<0.039**
	Control	36	72.0	14	28.0	
Platelet count	Cases	22	44.0	28	56.0	**<0.001**
	Control	49	98.0	1	2.0	

This table clearly shows the statistical significance of parameters namely serum prolactin, serum lactate dehydrogenase and platelet count between cases and controls as their p value are less than 0.05.

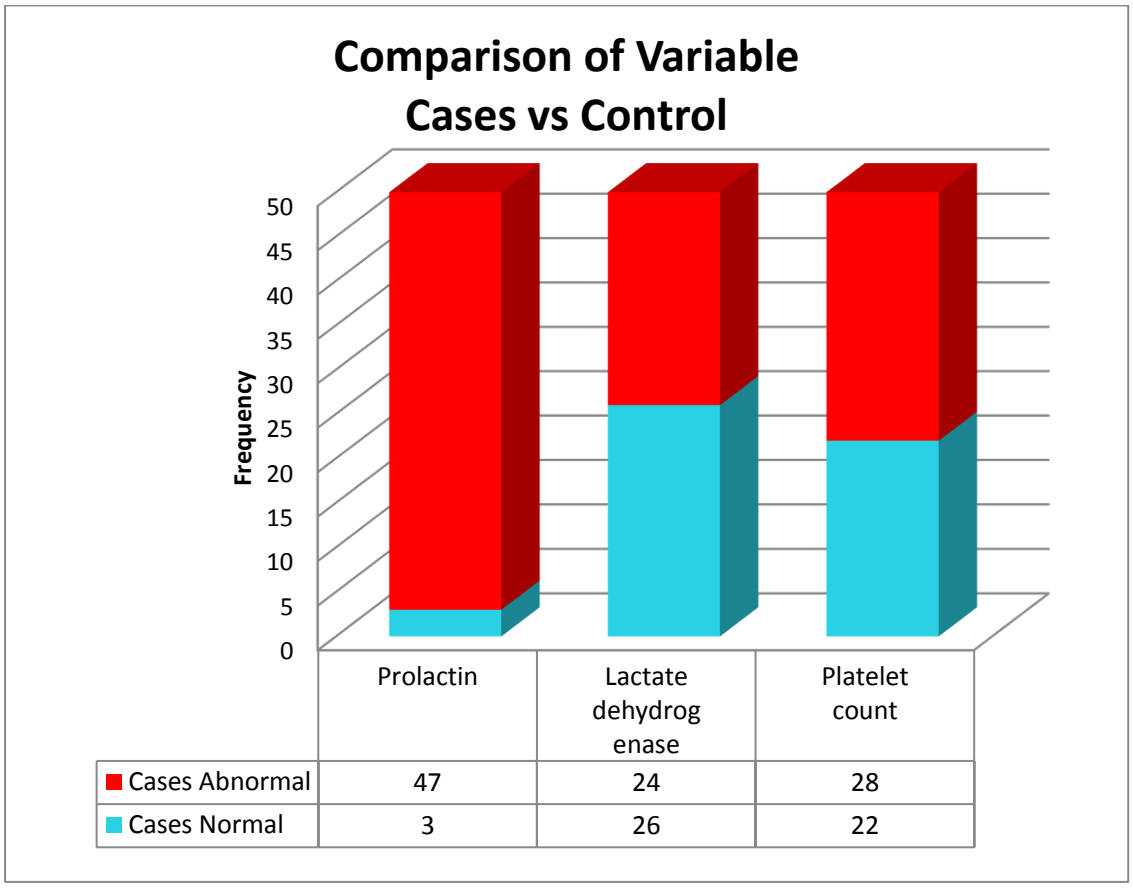
Serum Prolactin $p<0.001$ which is statistically significant.

Serum Lactate dehydrogenase $p<0.039$ which is statistically significant.

Serum Platelet count $p<0.001$ which is statistically significant.

FIGURE 27

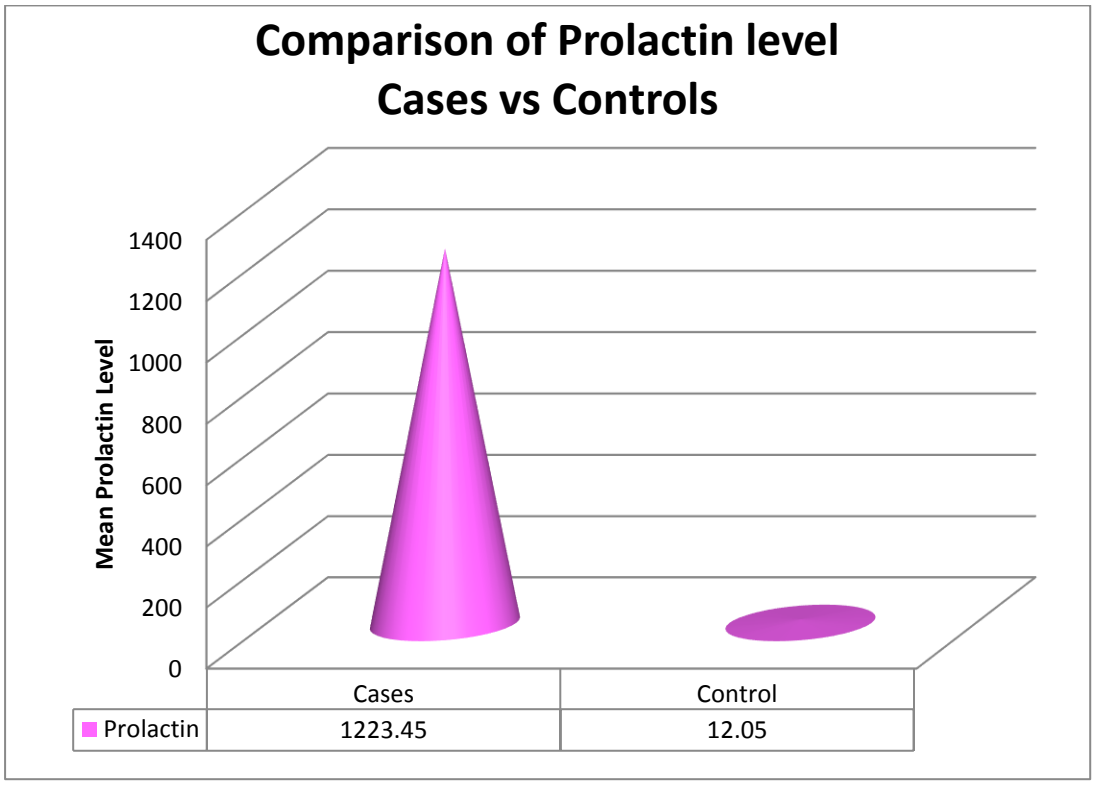
VARIABLE COMPRISION BETWEEN CASES AND CONTROL



The picture shows the degree of distribution of normal and abnormal among the cases population.

FIGURE 28

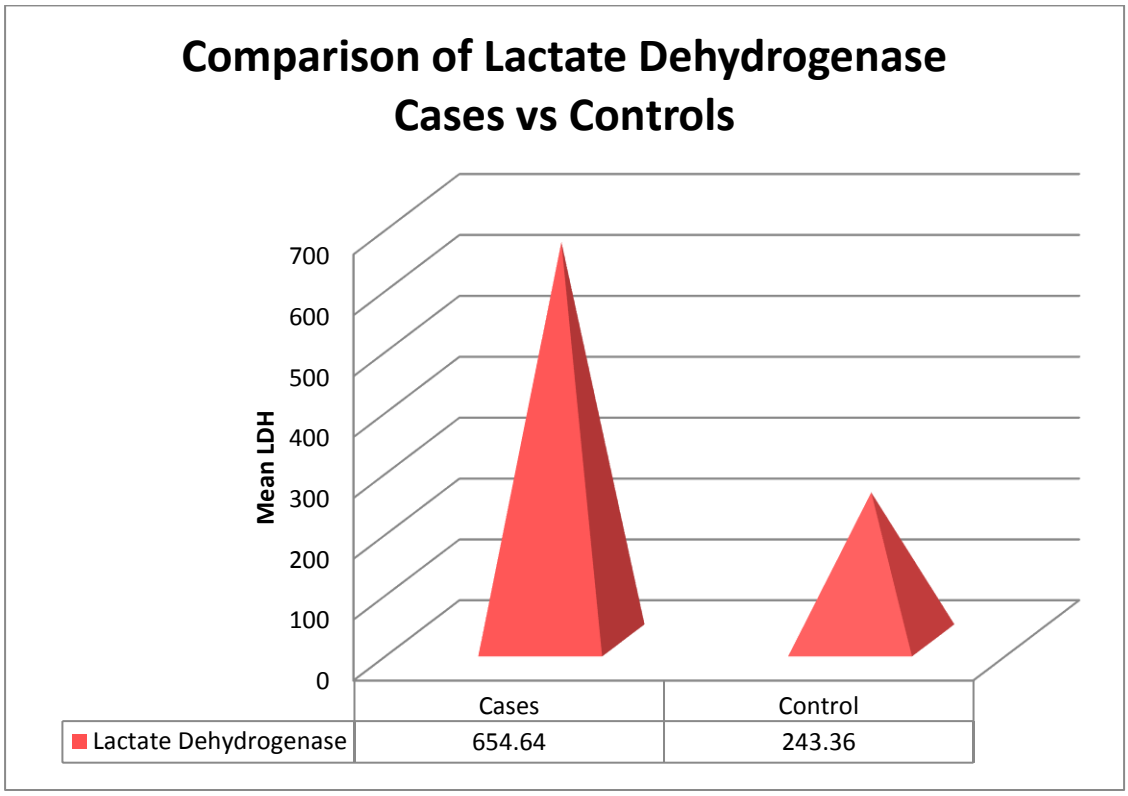
COMPARISION OF SERUM PROLACTIN LEVEL BETWEEN CASES AND CONTROLS



The picture shows comparison between cases and controls among serum prolactin levels. The mean Serum Prolactin concentration in cases is 1223.45 ng/ml.The mean Serum Prolactin in controls is 12.05 ng/ml.

FIGURE 29

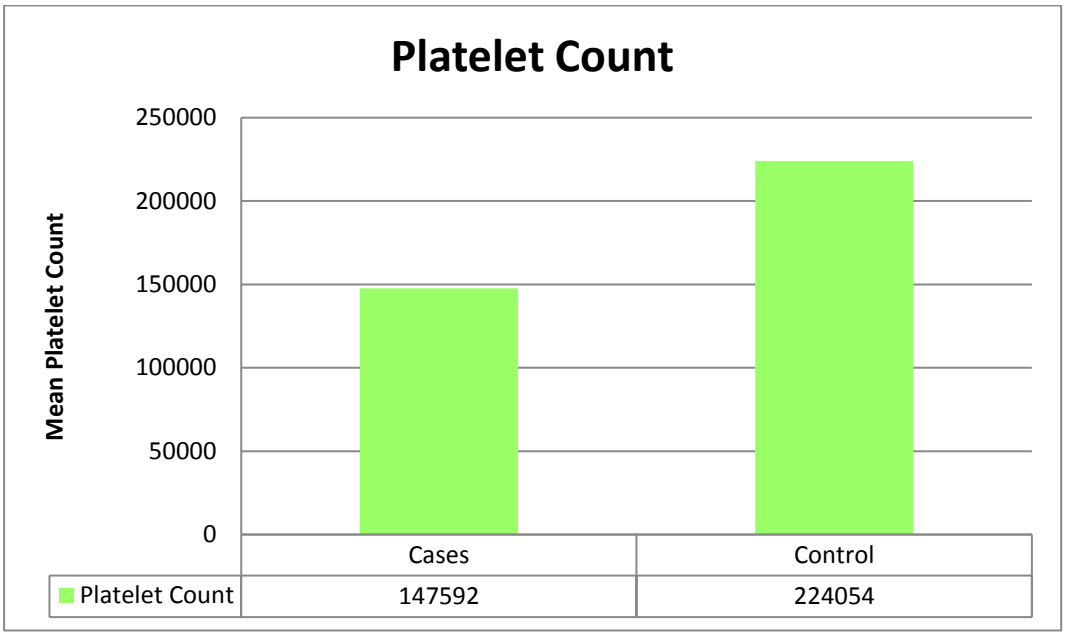
COMPARISION OF SERUM LACTATE DEHYDROGENASE LEVEL BETWEEN CASES AND CONTROLS



The picture shows comparison between cases and controls among serum Lactate dehydrogenase levels. The mean Serum Lactate Dehydrogenase concentration in cases is 654.64 IU/l. The mean Serum Lactate Dehydrogenase in controls is 243.36 IU/l.

FIGURE 30

COMPARISION OF PLATELET COUNT BETWEEN CASES AND CONTROLS



The picture shows comparison between cases and controls among Platelet count. The mean Platelet count in cases is 1,47,592 cells/cubic mm. The mean Platelet count in controls is 2,24,054 cells/cubic mm.

There is significant increase in mean platelet count in cases.

TABLE 11

Thyroid Profile	Group	Minimum	Maximum	Mean	Std. Deviation	P value
T3	Cases	0.7	2.1	1.40	0.41	0.418
	Controls	0.8	2.1	1.47	0.39	
T4	Cases	5.9	12.6	9.01	1.93	0.087
	Controls	6.4	12.4	9.60	1.45	
TSH	Cases	0.5	3.5	1.72	0.68	0.082
	Controls	0.5	2.5	1.52	0.47	

The table shows thyroid profile were in normal range between cases and controls.There is no significant difference as the p >0.05.

FIGURE 31

COMPARISION OF SERUM THYROID PROFILE BETWEEN CASES AND CONTROLS

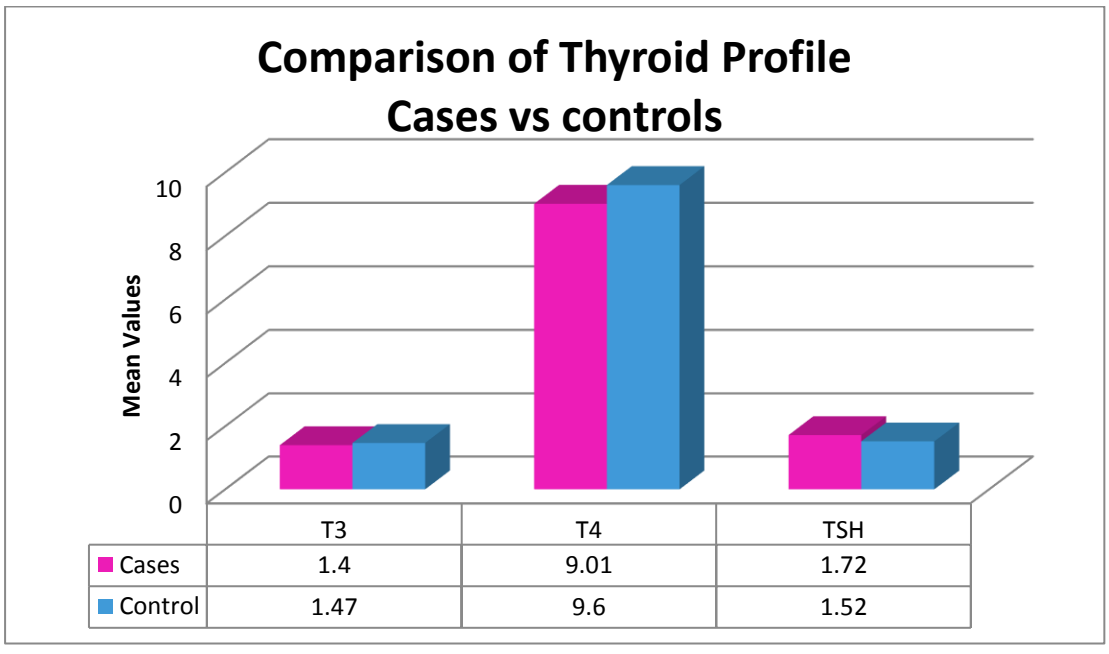


TABLE 12

Renal Function Test	Group	Normal		Abnormal		P value
		N	%	N	%	
Urea	Cases	49	98.0	1	2.0	**<0.001**
	Control	24	48.0	26	52.0	
Creatinine	Cases	13	26.0	37	74.0	**<0.001**
	Control	35	70.0	15	30.0	
Uric Acid	Cases	26	52.0	24	48.0	**<0.039**
	Control	36	72.0	14	28.0	
24 hours Urine Protein	Cases	3	6.0	47	94.0	**<0.001**
	Control	48	96.0	2	4.0	

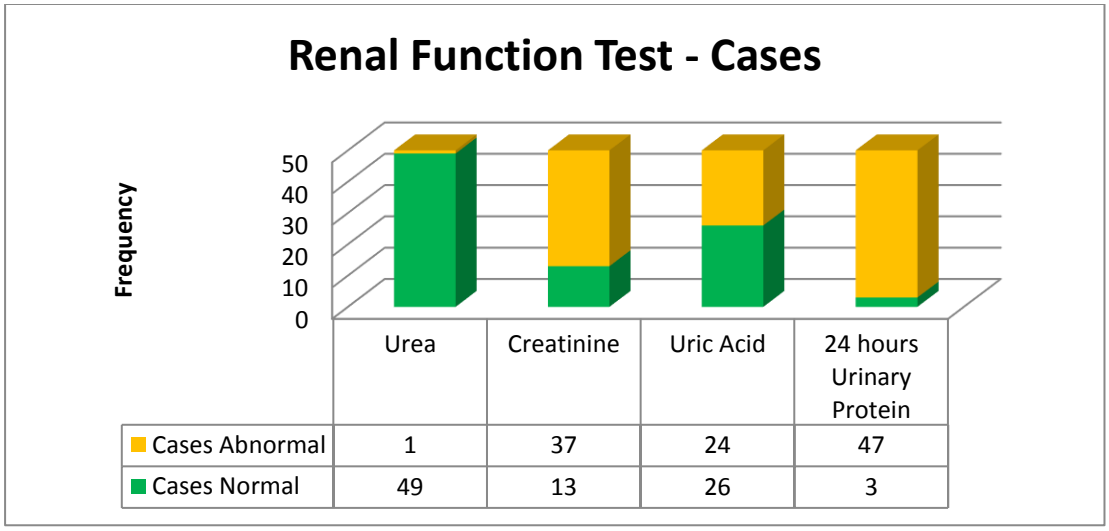
In renal function test, there is a significant difference between cases and controls in parameters urea, creatinine, uric acid and 24 hours urine protein as the p value is less than 0.05.

Serum urea and creatinine has $p<0.001$ which is statistically significant. Serum uric acid $p<0.039$ which is statistically significant. 24 hours Urine protein $p<0.01$ which is statistically significant.

The picture shows the distribution of normal and abnormal valuesin cases for the following parameters namely serum Urea, Creatinine, Uric acid and 24 hours urinary protein.

FIGURE 32

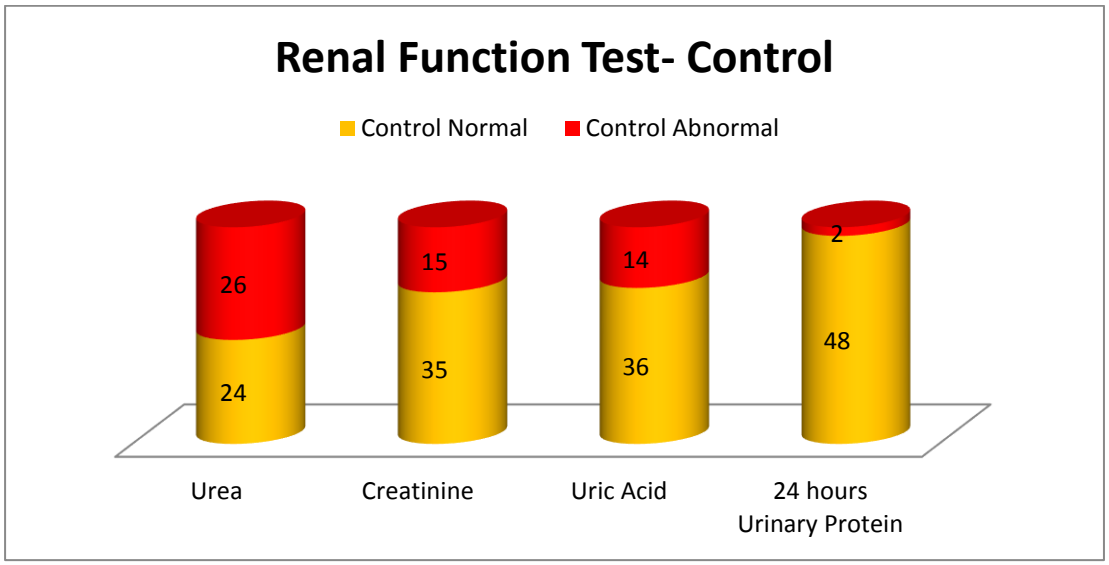
RENAL FUNCTION TEST PARAMETERS DISTRIBUTION AMONG CASES



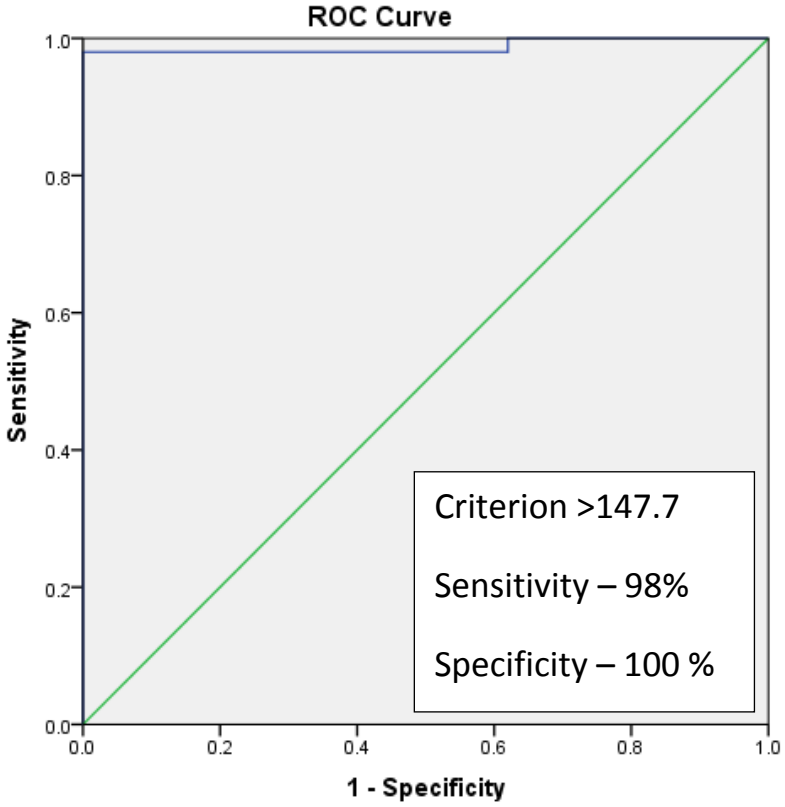
The picture shows the distribution of normal and abnormal values in controls for the following parameters namely serum Urea, Creatinine, Uric acid and 24 hours urinary protein.

FIGURE 33

RENAL FUNCTION TEST PARAMETERS DISTRIBUTION AMONG CONTROL



SERUM PROLACTIN- RECEIVER OPERATING CHARACTERISTIC CURVE



Area Under the Curve

Test Result Variable(s): PROLACTIN

Area	Std. Error[a]	Asymptotic Sig.[b]	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
.988	.012	.000	.963	1.000

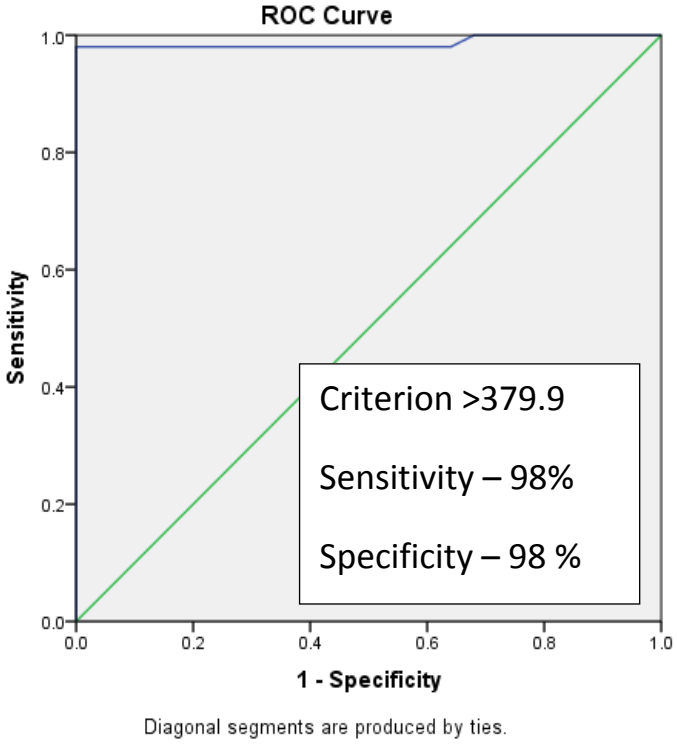
a. Under the nonparametric assumption

b. Null hypothesis: true area = 0.5

Remarks:

Sr. Prolactin >147.7 ng/mL is the predicting value for Preeclampsia, sensitivity is 98% and specificity is 100 %

SERUM LACTATE DEHYDROGENASE- RECEIVER OPERATING CHARACTERISTIC CURVE



Area Under the Curve

Test Result Variable(s): LACTATE_DEHYDROGENASE

Area	Std. Error[a]	Asymptotic Sig.[b]	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
.987	.013	.000	.961	1.000

The test result variable(s): LACTATEDEHYDROGENASE has at least one tie between the positive actual state group and the negative actual state group. Statistics may be biased.

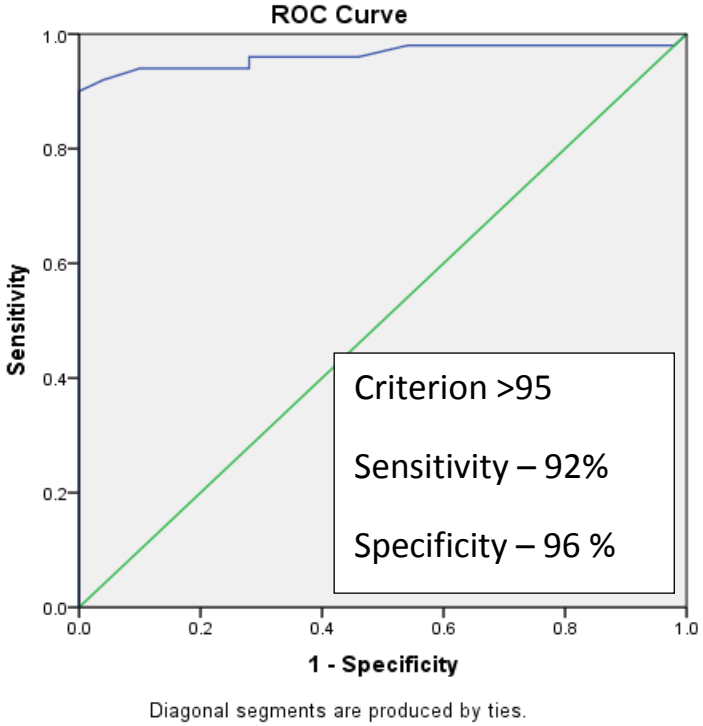
a. Under the nonparametric assumption

b. Null hypothesis: true area = 0.5

Remarks:

Sr. Lactate dehydrogenase >379.9 IU/L is the predicting value for Preeclampsia, sensitivity is 98% and specificity is 98 %

SERUM ALKALINE PHOSPHATASE- RECEIVER OPERATING CHARACTERISTIC CURVE



Area Under the Curve

Test Result Variable(s): ALKALINEPHOSPHATASE

Area	Std. Error[a]	Asymptotic Sig.[b]	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
.963	.023	.000	.918	1.000

The test result variable(s): ALKALINE_PHOSPHATASE has at least one tie between the positive actual state group and the negative actual state group. Statistics may be biased.

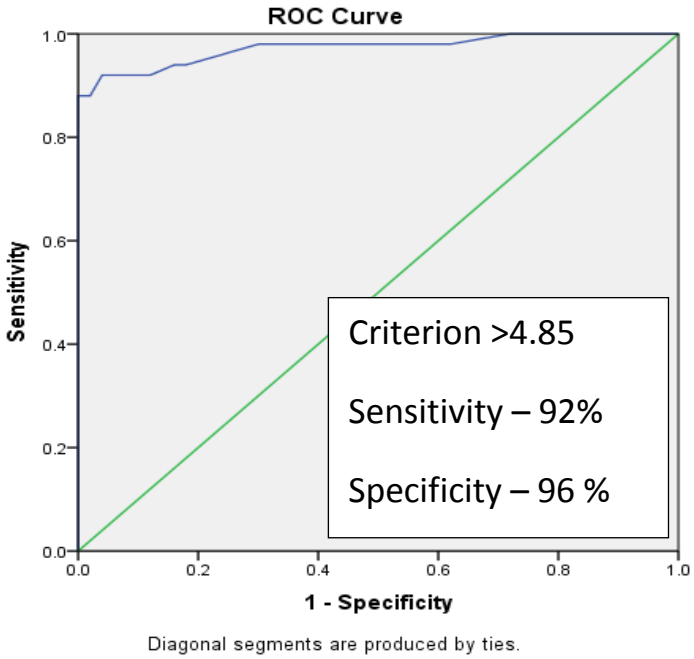
a. Under the nonparametric assumption

b. Null hypothesis: true area = 0.5

Remarks:

Sr. Alkaline phosphatase>95 IU/l is the predicting value for Preeclampsia, sensitivity is 92% and specificity is 96 %

SERUM URIC ACID- RECEIVER OPERATING CHARACTERISTIC CURVE



Area Under the Curve

Test Result Variable(s): URIC_ACID

Area	Std. Error[a]	Asymptotic Sig.[b]	Asymptotic 95% Confidence Interval	
			Lower Bound	Upper Bound
.973	.016	.000	.942	1.000

The test result variable(s): URIC_ACID has at least one tie between the positive actual state group and the negative actual state group. Statistics may be biased.

a. Under the nonparametric assumption

b. Null hypothesis: true area = 0.5

Remarks:

Sr. Uric acid>4.85 mg/dl is the predicting value for Preeclampsia, sensitivity is 92% and specificity is 96 %.

Among the four biochemical parameters that are statistically significant, serum Prolactin has a specificity 100% for preeclampsia.

Discussion

DISCUSSION

For a successful pregnancy, innumerous physiological adaptations occur in a woman's body, to facilitate good fetal growth, parturition and lactation. Any maladaptation in this causes serious maternal and fetal morbidity and mortality.

Recent research has thrown light on, FOAD hypothesis, i.e Fetal Origin Of Adult Disease, where low birth weight babies are linked to developing coronary artery disease, obesity, insulin resistance and hypertension in future. So, health care professionals are focused on implementing preventive measures. Preventive measures can be applied, only when high risk pregnancies are identified at an early stage. To identify high risk pregnancies, early reliable biomarkers are needed.

Among various obstetric disorders, pre eclampsia is known as "Disease of theories". Exact pathogenesis is still an enigma. Clinically the disease can be identified, but at a late stage and the only treatment available so far is to terminate the pregnancy. In order to provide a better fetal and maternal outcome, it becomes mandatory to identify this dreaded disease at a very earlier stage.

It is the need of the hour to focus on early reliable biomarkers. To solve this puzzle, various biochemical parameters are being haunted for decades, yet it is difficult to settle on one specific one.

In study, there is association of various biochemical parameters namely, serum prolactin, uric acid, alkaline phosphatase, lactate dehydrogenase and protein in urine.

History dates back to 1960's, when prolactin was associated with hypertension disorders in general population.

McGillivray et al, 1969 showed that both systolic and diastolic blood pressure falls significantly from that of first trimester in gestational weeks between 16-24.

In 1975, Horrobin et al, implicated that Prolactin has a possible role in the pathogenesis of pre eclampsia.

In 1975, Redman et al, showed higher prolactin levels in Pre eclampsia woman at 32 weeks compared to normal pregnancy, but later on it was shown that it could be also be due to methyl dopa drug intake.

In 1976, Biswas et al, showed negative correlation between prolactin and hypertensive pregnancies.

In 1977, Stumpe et al, and Lewis et al, showed the association of prolactin in hypertensive population was probably due to altered control of dopamine.

In the same year, Grant et al, proved the association of prolactin in hypertensive pregnancy was due to renal prostaglandin catabolism.

Since then, until date, various researches are done throughout entire world population both proving and disproving the above said hypothesis.

In this study, a total number of 100 antenatal mothers both from outpatient and inpatient side of obstetrics and gynecology department in government Coimbatore medical college hospital were studied.

In this study, among the various biochemical parameters done in Pre eclampsia mothers, there is significant association between serum prolactin and Pre eclampsia with significant p value <0.001.The mean prolactin value among controls is 12.5 ng/ml and cases is 1223.45 ng/ml. According to Receiver Operating Characteristic Curve, serum prolactin > 147.7 ng/ml is the predicting value for Pre eclampsia in this study, with sensitivity of 98% and specificity of 100%.

The specificity for serum prolactin in Pre eclampsia patients in this study is 100 %. This suggest that serum prolactin can be considered as a reliable marker for pre eclampsia.

Other important known biochemical parameters are serum uric acid, lactate dehydrogenase and alkaline phosphatase.

Robert et al, showed the importance of serum uric acid in gestational hypertension.

Lim LH et al, proved the clinical utility of measuring serum uric acid in hypertensive pregnancy.

Spencer et al, showed that apart from being a marker of pre eclampsia, it is also used as a marker for detecting aneuploidy in first trimester.

In this study, there is a positive association between serum uric acid and pre eclampsia as p value < 0.039.According to Receiver Operating Characteristic Curve, serum uric acid > 4.8 mg/dl is the predicting value for Pre eclampsia in this study, with sensitivity of 92% and specificity of 96%.

Bremme et al, showed that lactate dehydrogenase increases in pre eclampsia complicating pregnancy with liver failure and small for gestational age infants.

Jaiswar el al, proved significant increased levels of lactate dehydrogenase in Pre eclampsia and eclampsia.

In this study, there is significant association between serum lactate dehydrogenase and Pre eclampsia with significant p value <0.039. The mean serum lactate dehydrogenase value among controls is 243.36 IU/l and cases is 654.64 IU/l. According to Receiver Operating Characteristic Curve,

serum lactate dehydrogenase>379.9 IU/l is the predicting value for Pre eclampsia in this study, with sensitivity of 98% and specificity of 98%.

Hammoud et al, showed the association between increased serum alkaline phosphatase enzyme and HELLP syndrome in pre eclampsia.

Hunter et al, showed in his study that increased heat stable alkaline phosphatase is associated with pre eclampsia.

Eser et al, showed the increased liver enzymes including alkaline phosphatase in Pre eclampsia and the uses of plasma exchange in pre eclampsia.

In this study, there is significant association between serum alkaline phosphatase and Pre eclampsia with significant p value <0.001. According to Receiver Operating Characteristic Curve, serum alkaline phosphatase>95 IU/l is the predicting value for Pre eclampsia in this study, with sensitivity of 92% and specificity of 96%.

In this study, apart from the above-mentioned biochemical parameters, 24 hours urine protein has significant association with Pre eclampsia as its p value is < 0.01.

It is mandatory to conduct multiple studies in larger population to enable us to pick on early biomarkers in pre eclampsia.

Conclusion

CONCLUSION

- Pre eclampsia is the leading cause of fetal and maternal morbidity and mortality.
- To prevent the complications, it is necessary to subclassify Pre eclampsia based on biomarkers.
- Several research activities have analyzed different biomarkers in this area of expertise.
- In this study, there is significant association between Pre eclampsia and Serum Prolactin, Serum Uric Acid, Serum Lactate Dehydrogenase, Serum Alkaline Phosphatase and 24 hours urine protein.

Limitations of Study

LIMITATIONS OF THE STUDY

The study has following limitations;

- The sample is chosen from a community of population in and around Coimbatore and a small sample size of only 100 antenatal mothers.
- More precisely the Prolactin fragment namely 16KDa has to be quantified as this has proved to be associated with anti angiogenesis.
- Moreover, it is necessary to quantify a reference interval, above which it is capable of inducing hypertension and degree of disease severity.
- Achieving the above said goal, Pre eclampsia can be controlled by formulating drugs to inhibit prolactin 16 KDa, which remains to be investigated in future.

Bibliography

BIBLIOGRAPHY

1. Bell MJ. A Historical Overview of Preeclampsia-Eclampsia. J Obstet Gynecol Neonatal Nurs. 2010 Sep;39(5):510–8.

2. Website. History of Preeclampsia [Internet]. Preeclampsia Foundation Official Site. 2013 [cited 2019 Sep 14]. Available from: https://www.preeclampsia.org/history-of-preeclampsia/

3. Li R, Wang N, Xue M, Long W, Cheng C, Mi C, et al. A potential regulatory network among WDR86-AS1, miR-10b-3p, and LITAF is possibly involved in preeclampsia pathogenesis. Cell Signal. 2019 Mar 1;55:40–52.

4. Williams Obstetrics 25th Edition PDF FREE Download [Direct Link] [Internet]. Medicos Times. 2018 [cited 2019 Sep 16]. Available from: https://medicostimes.com/williams-obstetrics-pdf/

5. Rana Sarosh, Lemoine Elizabeth, Granger Joey, Karumanchi S. Ananth. Preeclampsia. Circ Res. 2019 Mar 29;124(7):1094–112.

6. Dekker G. The partner's role in the etiology of preeclampsia. J Reprod Immunol. 2002 Oct 1;57(1):203–15.

7. Williams PJ, Broughton Pipkin F. The genetics of pre-eclampsia and other hypertensive disorders of pregnancy. Best Pract Res Clin Obstet Gynaecol. 2011 Aug;25(4–4):405–17.

8. Michita RT, Kaminski V de L, Chies JAB. Genetic Variants in Preeclampsia: Lessons From Studies in Latin-American Populations. Front Physiol [Internet]. 2018 [cited 2019 Sep 10];9. Available from: https://www.frontiersin.org/articles/10.3389/fphys.2018.01771/full

9. Hui WWI, Chiu RWK. Noninvasive prenatal testing beyond genomic analysis: what the future holds. Curr Opin Obstet Gynecol. 2016 Apr;28(2):105–10.

10. Wang A, Rana S, Karumanchi SA. Preeclampsia: The Role of Angiogenic Factors in Its Pathogenesis. Physiology. 2009 Jun 1;24(3):147–58.

11. Helmo FR, Lopes AMM, Carneiro ACDM, Campos CG, Silva PB, dos Reis Monteiro MLG, et al. Angiogenic and antiangiogenic factors in preeclampsia. Pathol - Res Pract. 2018 Jan 1;214(1):7–14.

12. Powe CE, Levine RJ, Karumanchi SA. Preeclampsia, a disease of the maternal endothelium: the role of anti-angiogenic factors and implications for later cardiovascular disease. Circulation [Internet]. 2011 Jun 21 [cited 2019 Sep 22];123(24). Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3148781/

13. Redman CW, Sargent IL. Latest Advances in Understanding Preeclampsia. Science. 2005 Jun 10;308(5728):1592–4.

14. Morgan T, Craven C, Ward K. Human spiral artery renin-angiotensin system. Hypertens Dallas Tex 1979. 1998 Oct;32(4):683–7.

15. Seki H. The role of the renin–angiotensin system in the pathogenesis of preeclampsia – New insights into the renin–angiotensin system in preeclampsia. Med Hypotheses. 2014 Mar 1;82(3):362–7.

16. Pringle KG, Tadros MA, Callister RJ, Lumbers ER. The expression and localization of the human placental prorenin/renin-angiotensin system throughout pregnancy: roles in trophoblast invasion and angiogenesis? Placenta. 2011 Dec;32(12):956–62.

17. Goswami D, Tannetta DS, Magee LA, Fuchisawa A, Redman CWG, Sargent IL, et al. Excess syncytiotrophoblast microparticle shedding is a feature of early-onset pre-eclampsia, but not normotensive intrauterine growth restriction. Placenta. 2006 Jan;27(1):56–61.

18. Mincheva-Nilsson L, Baranov V. Placenta-derived exosomes and syncytiotrophoblast microparticles and their role in human reproduction: immune modulation for pregnancy success. Am J Reprod Immunol. 2014;72(5):440–57.

19. Sammar M, Drobnjak T, Mandala M, Gizurarson S, Huppertz B, Meiri H. Galectin 13 (PP13) Facilitates Remodeling and Structural Stabilization of Maternal Vessels during Pregnancy. Int J Mol Sci. 2019 Jan;20(13):3192.

20. Prolactin Expression and Secretion by Human Breast Glandular and Adipose Tissue Explants | The Journal of Clinical Endocrinology & Metabolism | Oxford Academic [Internet]. [cited 2019 Sep 9]. Available from: https://academic.oup.com/jcem/article/88/2/689/2845198

21. Reuwer AQ, Reuwer PJHM, van der Post JA, Cramer MJ, Kastelein JJP, Twickler MTB. Prolactin fragmentation by trophoblastic matrix metalloproteinases as a possible contributor to peripartum cardiomyopathy and pre-eclampsia. Med Hypotheses. 2010 Feb;74(2):348–52.

22. Saleem M, Martin H, Coates P. Prolactin Biology and Laboratory Measurement: An Update on Physiology and Current Analytical Issues. Clin Biochem Rev. 2018 Feb;39(1):3–16.

23. Transcriptional regulation by the helix bundle peptide hormones: growth hormone, prolactin, and hematopoietic cytokines. - PubMed - NCBI [Internet]. [cited 2019 Oct 2]. Available from: https://www.ncbi.nlm.nih.gov/pubmed/7843070

24. Redman CWG, Staff AC. Preeclampsia, biomarkers, syncytiotrophoblast stress, and placental capacity. Am J Obstet Gynecol. 2015 Oct 1;213(4):S9.e1-S9.e4.

25. Grattan DR, Jasoni CL, Liu X, Anderson GM, Herbison AE. Prolactin Regulation of Gonadotropin-Releasing Hormone Neurons to Suppress Luteinizing Hormone Secretion in Mice. Endocrinology. 2007 Sep;148(9):4344–51.

26. Grattan DR, Steyn FJ, Kokay IC, Anderson GM, Bunn SJ. Pregnancy-Induced Adaptation in the Neuroendocrine Control of Prolactin Secretion. J Neuroendocrinol. 2008;20(4):497–507.

27. Matrix metalloproteases from chondrocytes generate an antiangiogenic 16 kDa prolactin. - PubMed - NCBI [Internet]. [cited 2019 Oct 3]. Available from: https://www.ncbi.nlm.nih.gov/pubmed/16608881

28. Piwnica D, Touraine P, Struman I, Tabruyn S, Bolbach G, Clapp C, et al. Cathepsin D Processes Human Prolactin into Multiple 16K-Like N-Terminal Fragments: Study of Their Antiangiogenic Properties and Physiological Relevance. Mol Endocrinol. 2004 Oct 1;18(10):2522–42.

29. Bone Morphogenetic Protein Clips Angiogenic Role of Prolactin | Science Signaling [Internet]. [cited 2019 Oct 3]. Available from: https://stke.sciencemag.org/content/2007/391/tw221

30. Mustafa R, Ahmed S, Gupta A, Venuto RC. A Comprehensive Review of Hypertension in Pregnancy. J Pregnancy [Internet]. 2012 [cited 2019 Sep 12];2012. Available from: https://www.ncbi.nlm.nih.gov/pmc/articles/PMC3366228/

31. Triebel J, Bertsch T, Bollheimer C, Rios-Barrera D, Pearce CF, Hüfner M, et al. Principles of the prolactin/vasoinhibin axis. Am J Physiol-Regul Integr Comp Physiol. 2015 Aug 26;309(10):R1193–203.

32. Vasoinhibins: a family of N-terminal prolactin fragments that inhibit angiogenesis and vascular function. - PubMed - NCBI [Internet]. [cited 2019 Oct 3]. Available from: https://www.ncbi.nlm.nih.gov/pubmed/16809923

33. Lam C, Lim K-H, Kang D-H, Karumanchi SA. Uric acid and preeclampsia. Semin Nephrol. 2005 Jan;25(1):56–60.

34. Johnson RJ, Kanbay M, Kang D-H, Lozada LG-S, Feig D. Uric acid: A Clinically Useful Marker to Distinguish Preeclampsia from Gestational Hypertension. Hypertension. 2011 Oct;58(4):548–9.

35. Bainbridge SA, Roberts JM. Uric Acid as a Pathogenic Factor in Preeclampsia. Placenta. 2008 Mar;29(Suppl A):S67–72.

36. Staff AC, Benton SJ, von Dadelszen P, Roberts JM, Taylor RN, Powers RW, et al. Redefining preeclampsia using placenta-derived biomarkers. Hypertens Dallas Tex 1979. 2013 May;61(5):932–42.

37. He S, Bremme K, Kallner A, Blombäck M. Increased Concentrations of Lactate Dehydrogenase in Pregnancy with Preeclampsia: A Predictor for the Birth of Small-for-Gestational-Age Infants. Gynecol Obstet Invest. 1995;39(4):234–8.

38. Amirabi A, Danaii S. A Comparison of 4- and 24-Hour Urine Samples for the Diagnosis of Proteinuria in Pregnancy. Iran J Med Sci. 2011 Sep;36(3):167–71.

39. Kim MJ, Kim YN, Jung EJ, Jang HR, Byun JM, Jeong DH, et al. Is massive proteinuria associated with maternal and fetal morbidities in preeclampsia? Obstet Gynecol Sci. 2017 May;60(3):260–5.

PROFORMA

NAME: OP NO:

AGE/SEX:

ADDRESS: OCCUPATION:

DATE:

PRESENT HISTORY:

- H/o headache, blurring of vision, upper right abdominal pain
- H/o swelling of legs
- H/o pregnancy induced hypertension in previous pregnancy
- H/o any drug intake

PAST HISTORY:

- H/o liver/kidney/ Diabetes Mellitus/hypothyroid disease.
- H/o pregnancy induced hypertension in first degree relative

PERSONAL HISTORY:

- Attained Menarche at what age
- Age at marriage

ON EXAMINATION:

GENERAL EXAMINATION:

VITALS:

- PULSE RATE:
- Blood Pressure:
- BMI
- Cyanosis
- Icterus
- Pedal edema

Cardio vascular system:

Respiratory system

Per abdomen

Central nervous system

INVESTIGATIONS:

1. Serum prolactin- ELISA technique
2. Serum uric acid
3. Serum alkaline phosphatase
4. Liver function test

PATIENT CONSENT FORM

STUDY TITLE: A STUDY OF ASSOCIATION OF HIGH SERUM PROLACTIN LEVELS WITH OTHER BIOCHEMICAL PARAMETERS IN PRE ECLAMPSIA.

STUDY CENTRE: COIMBATORE MEDICAL COLLEGE HOSPITAL, COIMBATORE-18

PATIENT'S NAME: PATIENT'S AGE:

IDENTIFICATION NUMBER:

I confirm that I have understood the purpose and procedure of the above study. I have the opportunity to ask any questions and all my questions and doubts have been answered to my complete satisfaction.

I understand that my participation in this study is voluntary and that I am free to withdraw at any time without giving reason, without my legal rights being affected.

I understand that the sponsor of clinical study, working on sponsor's behalf, the ethical committee and the regulatory authorities will not need my permission to look at my health records, both in respect of the current study and any further research that may be conducted in relation to it, even if I withdraw from the study I agree to this access. However I understand that my identity would not be revealed in any information released to third parties unless as required under the law. I agree not to restrict the use of any data or results that arise from the study.

I hereby consent to participate in this study.

I hereby give permission to undergo complete clinical examination and diagnostic tests including hematological, biochemical and radiological tests.

Signature/thumb impression
Patient's name and address
Signature of the investigator:
Name of the investigator:

Place: Place:

Date: Date:

ஒப்புதல்படிவம்

நோயாளியின் பெயர்:

பாலினம் : வயது:

பெற்றோர் பெயர் :

முகவரி :

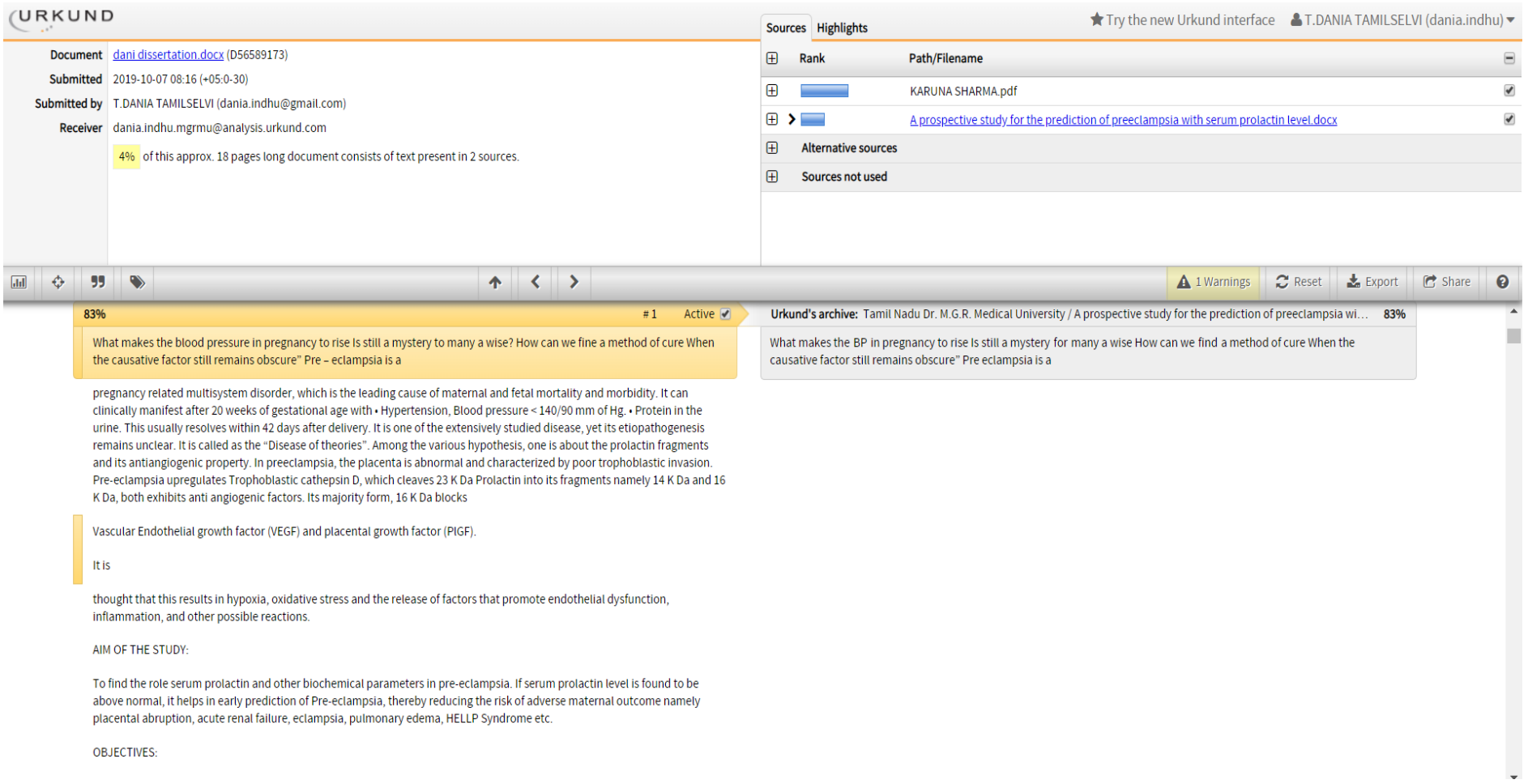
அரசு கோவை மருத்துவக் கல்லூரியில் **உயிர் வேதியல்** துறையில் பட்ட மேற்படிப்பு பயிலும் மாணவி மரு. **த.டானியா தமிழ்ச்செல்வி அவர்கள் மேற்கொள்ளும் முன்கூல்வலிப்பு உள்ள மகப்பேறு தாய்மார்களின் ரத்தத்தில் பிரோலேக்ட்டின் அளவை** பற்றிய ஆய்வில் செய்முறை மற்றும் அனைத்து விளக்கங்களையும் கேட்டுக் கொண்டு எனது சந்தேகளை தெரிவுபடுத்திக் கொண்டேன் என்பதை தெரிவித்துக் கொள்கிறேன்.

இந்த ஆய்வில் நான் முழு சம்மதத்துடனும், சுயசிந்தனையுடனும் கலந்து கொள்ள சம்மதிக்கிறேன்.

இந்த ஆய்வில் என்னைப் பற்றிய அனைத்து விவரங்கள் பாதுகாக்கப்படுவதுடன் இதன் முடிவுகள் ஆய்விதழில் வெளியிடப்படுவதில் ஆட்சேபனை இல்லை என்பதை தெரிவித்துக்கொள்கிறேன். எந்த நேரத்திலும் இந்த ஆய்விலிருந்து நான் விலகிக் கொள்ள எனக்கு உரிமை உண்டு என்பதையும் அறிவேன்.

இடம் :

தேதி : கையொப்பம் / ரேகை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What makes the blood pressure in pregnancy to rise Is still a mystery to many a wise? How can we fine a method of cure When the causative factor still remains obscure" Pre – eclampsia is a
pregnancy related multisystem disorder, which is the leading cause of maternal and fetal mortality and morbidity. It can clinically manifest after 20 weeks of gestational age with • Hypertension, Blood pressure < 140/90 mm of Hg. • Protein in the urine. This usually resolves within 42 days after delivery. It is one of the extensively studied disease, yet its etiopathogenesis remains unclear. It is called as the "Disease of theories". Among the various hypothesis, one is about the prolactin fragments and its antiangiogenic property. In preeclampsia, the placenta is abnormal and characterized by poor trophoblastic invasion. Pre-eclampsia upregulates Trophoblastic cathepsin D, which cleaves 23 K Da Prolactin into its fragments namely 14 K Da and 16 K Da, both exhibits anti angiogenic factors. Its majority form, 16 K Da blocks
Vascular Endothelial growth factor (VEGF) and placental growth factor (PlGF).
It is
thought that this results in hypoxia, oxidative stress and the release of factors that promote endothelial dysfunction, inflammation, and other possible reactions.
AIM OF THE STUDY:
To find the role serum prolactin and other biochemical parameters in pre-eclampsia. If serum prolactin level is found to be above normal, it helps in early prediction of Pre-eclampsia, thereby reducing the risk of adverse maternal outcome namely placental abruption, acute renal failure, eclampsia, pulmonary edema, HELLP Syndrome etc.
OBJECTIVES:
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What makes the BP in pregnancy to rise Is still a mystery for many a wise How can we find a method of cure When the causative factor still remains obscure" Pre eclampsia is a

MASTER CHART

S_NO	TYPE	AGE	SEX	PROLACTIN	URIC_ACID	LACTATE_ DEHYDROGENASE	ALKALINE_ PHOSPHATASE	T3	T4	TSH	BILIRUBIN_TOTAL	BILIRUBIN_ DIRECT	AST	ALT	PROTEIN	ALBUMIN	GLOBULIN	UREA	CREATININE	24_HOURS_ URINE_ PROTEIN	PLATELET_ COUNT
1	1	29	1	968.9	7.2	1029	157	1.3	7.4	2.1	0.9	0.4	42	38.2	6.3	3.5	2.8	20.3	0.86	0.42	124000
2	1	26	1	564.9	6.9	550	127	2	11.1	1.9	0.84	0.32	37	27.6	6.8	3.9	2.9	31.2	0.92	0.39	214000
3	1	31	1	270.3	6.2	488	151	0.7	6.2	3.5	0.73	0.39	29	18.9	6.7	3.8	2.9	28.3	0.63	0.27	256000
4	1	31	1	2000	8.1	592	152	0.9	5.9	2.6	1.3	0.6	92	82.6	6.1	3	3.1	26.5	0.58	0.56	101300
5	1	23	1	386.1	5.9	625	48	2.1	10.9	1.7	1	0.4	27	30.2	7.1	4.2	2.9	23.4	0.45	0.23	241000
6	1	27	1	2000	7.8	521	140	1.6	8.4	2.7	0.65	0.38	84	64.8	6.9	3.8	3.1	19.6	0.68	0.52	93000
7	1	23	1	581.1	6.3	668	129	1.5	7.8	1.7	0.82	0.29	73	59.3	6.7	3.5	3.2	22.5	0.49	0.39	214000
8	1	21	1	2000	8.1	519	159	0.9	6.2	2.9	1.1	0.5	65	87.1	6.5	3.5	3	31.7	0.67	0.67	95000
9	1	30	1	1885	7.2	564	145	0.9	5.9	3.2	0.97	0.37	56	48.2	6.6	3.8	2.8	36.4	0.53	0.61	102000
10	1	21	1	399	4.9	218	185	2	11.8	0.8	0.76	0.31	33	29.4	6.8	3.4	3.4	29.4	0.84	0.41	219000
11	1	23	1	766	5.3	464	136	1.4	7.9	1.6	0.84	0.28	40	39.7	6.4	3.8	2.6	33.5	1.1	0.52	193000
12	1	28	1	2000	7.9	992	134	1.9	10.3	1.9	1.3	0.6	97	85.1	6.2	2.9	3.3	28.7	0.63	0.58	98000
13	1	22	1	597.1	6.2	766	132	0.9	6.7	2.6	0.94	0.38	18	12.8	7.1	3.8	3.3	23	0.28	0.48	230000
14	1	24	1	1956	7.2	914	137	1.3	8.3	2.5	0.68	0.39	89	76.1	6.9	3.7	3.2	31.8	0.86	0.59	217000
15	1	26	1	2000	8.5	620	168	1.8	12.1	0.8	1.23	0.52	103	138	6.3	3.8	2.5	26.8	0.57	0.81	93000
16	1	17	1	481.9	4.6	635	69	1.4	8.9	1.6	0.97	0.41	62	59.3	6.8	3.4	3.4	22.4	0.63	0.37	201000
17	1	25	1	2000	8.6	724	148	1.12	9.4	2.6	0.97	0.39	98	86.5	6.4	3.8	2.6	19.7	0.52	0.41	103400
18	1	26	1	1985	7.5	639	225	0.9	6.8	1.4	0.84	0.38	90	74.6	7.5	4.8	2.7	23.5	0.58	0.58	153000
19	1	29	1	2000	8.1	527	156	1.9	9.2	1.1	0.76	0.37	98	98.3	6.9	3.8	3.1	28.6	0.68	0.51	99100
20	1	26	1	896.8	6.5	588	149	1.5	10.7	0.6	0.85	0.36	40	67.1	6.5	3.7	2.8	22.7	0.45	0.43	176000
21	1	20	1	2000	7.2	712	179	0.8	5.9	1.2	1.1	0.4	105	98.3	6.3	3.9	2.4	29.8	0.36	0.62	94000
22	1	19	1	681.6	4.1	625	152	1.2	9.6	1.9	0.94	0.41	35	31.9	7.2	4.6	2.6	31.4	0.84	0.45	173000

S_NO	TYPE	AGE	SEX	PROLACTIN	URIC_ACID	LACTATE_DEHYDROGENASE	ALKALINE_PHOSPHATASE	T3	T4	TSH	BILIRUBIN_TOTAL	BILIRUBIN_DIRECT	AST	ALT	PROTEIN	ALBUMIN	GLOBULIN	UREA	CREATININE	24_HOURS_URINE_PROTEIN	PLATELET_COUNT
23	1	22	1	399.6	5.4	535	151	1.8	10.3	0.9	0.84	0.39	36	28.6	6.5	3.6	2.9	28.6	0.46	0.38	102000
24	1	19	1	663.9	5.3	466	232	2.1	12.1	0.5	0.76	0.41	29	21.6	6.8	3.5	3.3	31.8	0.84	0.45	218000
25	1	24	1	2000	8.3	472	203	0.9	6.8	1.7	0.82	0.39	114	98.3	6.9	3.9	3	29.6	0.36	0.61	89500
26	1	29	1	987.8	6.2	1153	166	1.1	9.1	2.1	0.79	0.41	57	43.6	6.9	3.8	3.1	28.4	0.48	0.57	143000
27	1	18	1	597.1	5.8	1098	150	1.6	7.9	1.3	0.68	0.38	22	20.3	7.5	4.5	3	25.3	0.68	0.32	206000
28	1	26	1	483.9	4.9	544	163	1.9	10.8	2.1	0.76	0.36	64	56.8	6.4	3.9	2.5	26.8	0.35	0.37	182000
29	1	25	1	796.9	6.8	840	174	1.4	9.7	1.1	0.82	0.41	39	32.4	6.8	3.3	3.5	31.4	0.46	0.49	114000
30	1	20	1	2000	8.2	477	191	1.2	10.1	1.7	1.3	0.62	88	104	6.7	3.8	2.9	29.4	0.57	0.68	106000
31	1	22	1	1882	7.6	1027	157	0.9	5.9	2.1	1.1	0.51	113	97.2	7.1	4.2	2.9	28.3	1	0.52	97000
32	1	21	1	984	7.2	594	119	1.1	7.5	1.8	0.98	0.36	76	63.5	6.8	3.8	3	22.7	0.39	0.32	186000
33	1	23	1	380	5.3	737	94	2	12.1	0.7	0.76	0.41	48	41.5	7.6	4.3	3.3	26.9	0.31	0.41	210000
34	1	20	1	2000	8.2	635	191	1.1	7.3	1.8	0.9	0.4	104	89.6	5.9	3.6	2.3	23.6	0.28	0.64	96000
35	1	20	1	1363	7.1	735	173	1.4	9.6	1.5	0.89	0.31	115	97.3	6.6	3.8	2.8	24.1	0.34	0.59	106000
36	1	24	1	657.1	5.6	970	151	1.8	12.1	1.1	1.1	0.52	68	56.8	6.2	3.7	2.5	20.3	0.52	0.43	149000
37	1	29	1	498	4.2	585	96	2.1	8.3	2.1	0.98	0.36	62	54.3	7.2	4.2	3	30.8	0.38	0.38	207000
38	1	30	1	1836	8.5	671	192	1.6	9.1	1.4	0.76	0.41	103	98.2	6.3	3.8	2.5	25.6	0.61	5.7	101000
39	1	30	1	675	6.1	675	213	1.1	10.4	0.9	0.87	0.38	84	76.3	6.5	3.8	2.7	23.6	0.93	0.32	135000
40	1	18	1	982	6.4	496	168	1.8	9.8	1.1	0.94	0.39	39	28.1	6.9	3.6	3.3	27.1	0.28	0.49	172000
41	1	31	1	1728	7.6	637	158	1.4	10.3	2.3	0.68	0.41	88	77.5	6.5	3.8	2.7	30.9	1.1	0.63	91000
42	1	24	1	2000	8.3	603	154	1	8.9	1.6	1.21	0.6	106	98.3	7.1	4.2	2.9	28.1	0.38	0.72	98000
43	1	26	1	1453	7.9	617	153	1.6	9.6	1.1	1.1	0.42	84	78.6	6.8	3.9	2.9	30.7	49	0.53	103000
44	1	20	1	1890	6.8	489	223	1.5	7.9	1.4	1.31	0.53	84	72.1	6.9	3	3.9	23.1	0.73	0.57	127000
45	1	28	1	2000	8.5	491	181	0.9	6.3	2.7	0.98	0.37	108	96.8	5.8	3.2	2.6	28.6	0.91	0.69	98300
46	1	21	1	356	6.1	459	133	1.9	12.1	1.6	0.85	0.48	82	68.3	6.9	3.6	3.3	21.4	0.58	0.37	203000

S_NO	TYPE	AGE	SEX	PROLACTIN	URIC_ACID	LACTATE_ DEHYDROGENASE	ALKALINE_ PHOSPHATASE	T3	T4	TSH	BILIRUBIN_TOTAL	BILIRUBIN_ DIRECT	AST	ALT	PROTEIN	ALBUMIN	GLOBULIN	UREA	CREATININE	24_HOURS_ URINE_ PROTEIN	PLATELET_ COUNT
47	1	32	1	762	5.3	677	177	1.6	12.6	1.3	0.76	0.39	65	50.7	6.8	3.7	3.1	22.8	0.61	0.46	184000
48	1	28	1	2000	8.1	948	185	0.7	8.6	1.8	0.84	0.37	109	98.6	5.9	3.4	2.5	26.4	0.53	0.64	98000
49	1	22	1	1368	5.4	686	279	1.1	9.2	2.1	0.92	0.35	100	84.1	6.8	4.1	2.7	28.4	0.64	0.39	112000
50	2	26	1	9.6	3.6	435	85	1.6	8.9	1.7	0.3	0.12	26	18.4	6.5	4.1	2.4	12.2	0.78	0.015	156000
51	2	22	1	10.7	3.9	235	67	1.2	7.9	1.5	0.64	0.25	13	11.6	6.9	4	2.9	19.1	1.06	0.03	183000
52	2	27	1	16.3	4.1	318	96	1.7	10.4	2.4	0.45	0.19	15	3.7	7.4	3.8	3.6	10.7	0.94	0.01	201000
53	2	28	1	25.1	3.8	287	78	1	6.4	1.4	0.51	0.22	14	11	7.5	5	2.5	11.7	0.88	0.025	196000
54	2	19	1	7.9	3.2	309	68	0.9	8.3	1.9	0.42	0.19	12	12.2	7.5	4.4	3.1	11	0.75	0.21	138000
55	2	21	1	12.9	3.4	405	53	2.1	12.4	0.5	0.51	0.2	13	12.2	7.2	4.9	2.3	17.3	1.02	0.03	176000
56	2	20	1	6.1	4.7	296	69	0.8	8.7	1.3	0.82	0.43	25	19.3	7.7	4.5	3.2	25	0.7	0.05	185000
57	2	24	1	18.7	3.6	304	91	1.4	10.3	1.7	0.37	0.15	17	19.8	7.3	4.1	3.2	19.8	0.79	0.02	197000
58	2	20	1	22.2	2.9	158	86	1.3	9.5	1.1	0.78	0.37	21	28	7.5	4.9	2.6	23.5	0.87	0.16	207000
59	2	32	1	4.9	3.8	306	62	1.91	11.8	2.1	0.23	0.1	25	14.1	7.3	4.3	3	14.8	0.89	0.21	186000
60	2	28	1	11.3	4.6	267	93	2	12.3	0.9	0.47	0.19	29	18	7.5	4.5	3	17.2	1.12	0.18	207000
61	2	20	1	5.2	3.8	168	54	1.9	10.6	1.3	0.14	0.07	87	42.8	5.6	2.9	2.7	18.9	0.94	0.14	257000
62	2	31	1	10.9	3.9	203	68	1.12	10.9	1.6	0.2	0.09	11	12.2	6.8	3.2	3.6	30.3	0.88	0.2	204000
63	2	28	1	8.7	4.7	217	76	1.5	9.8	1.1	0.44	0.18	30	28.1	7.3	4.3	3	27.3	0.73	0.18	269000
64	2	26	1	13.9	2.8	307	48	1.9	11.7	0.8	0.33	0.15	36	23.9	7.6	4.9	2.7	13	1.1	0.16	198000
65	2	20	1	17.3	3.8	205	69	1.2	10.4	1.6	0.47	0.19	14	11	7	4.2	2.8	12.8	0.65	0.15	176000
66	2	30	1	12.7	3.9	257	63	1.4	10.4	1.2	0.18	0.1	36	58.7	6.7	4.8	1.9	18.5	0.76	0.21	195000
67	2	19	1	7.3	3.5	301	88	1.1	9.6	1.9	0.34	0.14	19	24.3	6.6	3.1	3.5	15.1	0.85	0.16	257000
68	2	25	1	14	4.9	268	94	2.1	12	0.7	0.17	0.09	10	11.9	6.7	3	3.7	12	0.94	0.2	230000
69	2	22	1	17.2	3.8	124	68	1.9	8.4	1.2	0.71	0.24	19	13.6	6.7	3	3.7	16	0.83	18	273000
70	2	21	1	14.1	3.6	306	87	1.7	7.4	2.5	0.4	0.13	21	19	4.6	1.9	2.7	29.2	0.97	0.1	218000

S_NO	TYPE	AGE	SEX	PROLACTIN	URIC_ACID	LACTATE_ DEHYDROGENASE	ALKALINE_ PHOSPHATASE	T3	T4	TSH	BILIRUBIN_TOTAL	BILIRUBIN_ DIRECT	AST	ALT	PROTEIN	ALBUMIN	GLOBULIN	UREA	CREATININE	24_HOURS_ URINE_ PROTEIN	PLATELET_ COUNT
71	2	28	1	18.4	4.7	296	59	1.5	10.2	1.6	0.22	0.11	15	10.7	7.4	4	3.4	21.1	1.18	16	196000
72	2	26	1	13.1	3.8	354	67	1.6	9.7	1.2	0.86	0.38	15	10.7	6	2.7	3.3	9.6	0.84	0.12	193000
73	2	22	1	11.8	3.9	168	94	1.3	11	1.7	0.46	0.2	14	10.1	7.4	4.1	3.3	18	0.81	0.17	205000
74	2	20	1	7.9	4.6	329	62	1.5	7.4	1.9	0.5	0.21	16	8.3	5.6	2.8	2.8	12.2	0.76	0.21	284000
75	2	23	1	5.7	3.6	203	96	1.1	9.2	1.1	1.28	0.42	34	17.8	5.7	2.8	2.9	16.5	0.98	0.08	263000
76	2	30	1	9.4	3.5	250	84	2	11.1	0.7	1.14	0.85	205	94.7	3.5	1.7	1.8	19.4	0.65	0.06	274000
77	2	29	1	11.7	4.8	167	57	1.9	9.2	1.7	0.54	0.19	18	12	8	4.7	3.3	18.5	1.15	0.27	196000
78	2	22	1	14.6	3.8	218	83	1.2	8.8	1.4	0.4	0.1	17	17.7	5.5	3.1	2.4	12.9	0.56	0.15	276000
79	2	27	1	18.2	3.5	159	64	0.9	7.3	2.1	0.4	0.1	17	13.9	7.6	4.4	3.2	19.6	0.98	0.24	283000
80	2	25	1	16.9	2.9	230	92	1.5	9.1	1.4	1.2	0.3	17	12.3	4.3	2.1	2.2	33.4	0.88	0.13	294000
81	2	30	1	12.5	3.6	218	63	0.8	6.9	1.1	0.4	0.13	16	17.8	6.9	4.9	2	28.4	0.94	0.18	187000
82	2	28	1	18.3	5.2	234	84	1.6	10.2	1.4	0.19	0.07	12	20.1	6.5	4.4	21	11.1	0.54	0.1	201000
83	2	26	1	8.7	4.2	251	59	1.2	9.1	2.1	0.5	0.15	20	21	5	2.7	2.3	14.7	0.58	0.24	195000
84	2	24	1	10.4	3.8	183	87	1	7.9	1.2	0.57	0.21	28	24.1	6.4	3	3.4	16.5	0.69	0.19	198000
85	2	29	1	6.2	4.1	238	69	1.9	10.2	1.8	0.33	0.15	12	17.6	6.9	4	2.9	10.3	0.67	0.16	203000
86	2	22	1	9.4	3.8	206	86	1.7	9.2	1.3	3.76	1.23	28	18.4	6.2	3.3	2.9	13.8	0.61	0.21	201700
87	2	21	1	16.7	3.5	228	94	2.1	11.4	0.8	0.3	0.11	22	19.9	6.4	3.4	3	18	0.57	0.01	198000
88	2	27	1	9.4	4.3	230	83	0.9	10.1	1.3	1.01	0.37	29	66.2	7.9	4.1	3.8	19.2	0.7	0.21	230000
89	2	20	1	10.2	3.8	201	82	1.2	9.2	1.4	0.43	0.07	18	24.8	6.7	3.8	2.9	14.3	0.55	0.17	201000
90	2	31	1	18.1	3.4	270	69	1	7.3	2.1	0.3	0.14	29	24.1	7.5	5.7	2.8	21.1	0.59	0.1	254000
91	2	23	1	6.4	4.2	238	67	1.8	11.3	1.9	0.14	0.08	17	20.9	7.5	4.2	3.3	14.8	0.61	0.02	286000
92	2	20	1	3.4	3.8	164	84	0.9	9.5	2.1	0.29	0.14	76	86.3	6.9	4.3	2.6	24.9	0.73	0.17	271000
93	2	24	1	16.3	3.5	221	80	1.6	10.1	1.7	0.24	0.14	16	18.1	6.8	4	2.8	29.5	0.77	0.24	204000
94	2	21	1	17.2	3.4	237	59	1.5	8.7	1.3	0.21	0.11	18	13.9	7.2	4.5	2.7	24.1	0.73	0.13	208000

S_NO	TYPE	AGE	SEX	PROLACTIN	URIC_ACID	LACTATE_ DEHYDROGENASE	ALKALINE_ PHOSPHATASE	T3	T4	TSH	BILIRUBIN_TOTAL	BILIRUBIN_ DIRECT	AST	ALT	PROTEIN	ALBUMIN	GLOBULIN	UREA	CREATININE	24_HOURS_ URINE_ PROTEIN	PLATELET_ COUNT
95	2	28	1	10.1	3.2	308	87	1.9	9.5	1.8	0.41	0.2	12	15.6	7.4	4.5	2.9	11.6	0.62	0.17	203000
96	2	20	1	9.2	4.1	238	63	2.1	11.7	2.4	0.41	0.12	16	30.7	7.2	4.5	2.7	12.3	0.7	0.05	286000
97	2	22	1	11.3	3.8	207	64	1.3	8.9	1.2	0.52	0.21	22	17.6	6.9	3.7	3.2	14.3	0.61	0.16	253000
98	2	28	1	8.5	3.2	239	57	1.2	9.6	1.9	0.18	0.1	12	13.7	6.9	3.8	3.1	13.4	0.62	0.04	291000
99	2	26	1	6.7	4.2	134	68	1.5	8.3	1.7	0.3	0.3	16	21.8	7.2	4.8	2.4	19.3	0.71	0.1	239000
100	2	24	1	8.4	3.6	308	59	1.7	9.1	2.1	0.48	0.17	16	19.4	6.8	3.9	2.9	20.7	0.91	0.16	276000

FIGURE 2:

WORLD MAP SHOWING PREVALENCE OF PRE ECLAMPSIA

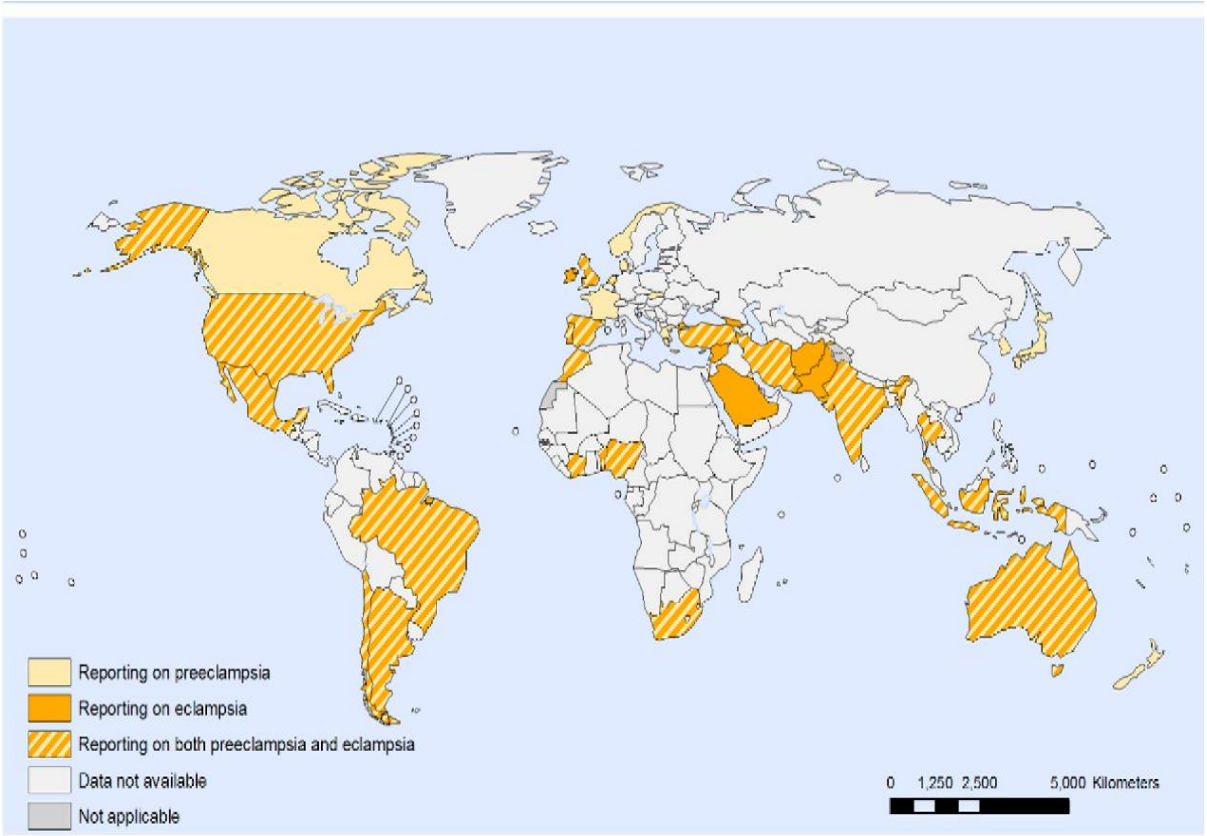


FIGURE 3:

AN INTEGRATED PICTURE OF KEY EVENTS IN PRE ECLAMPSIA.[8]

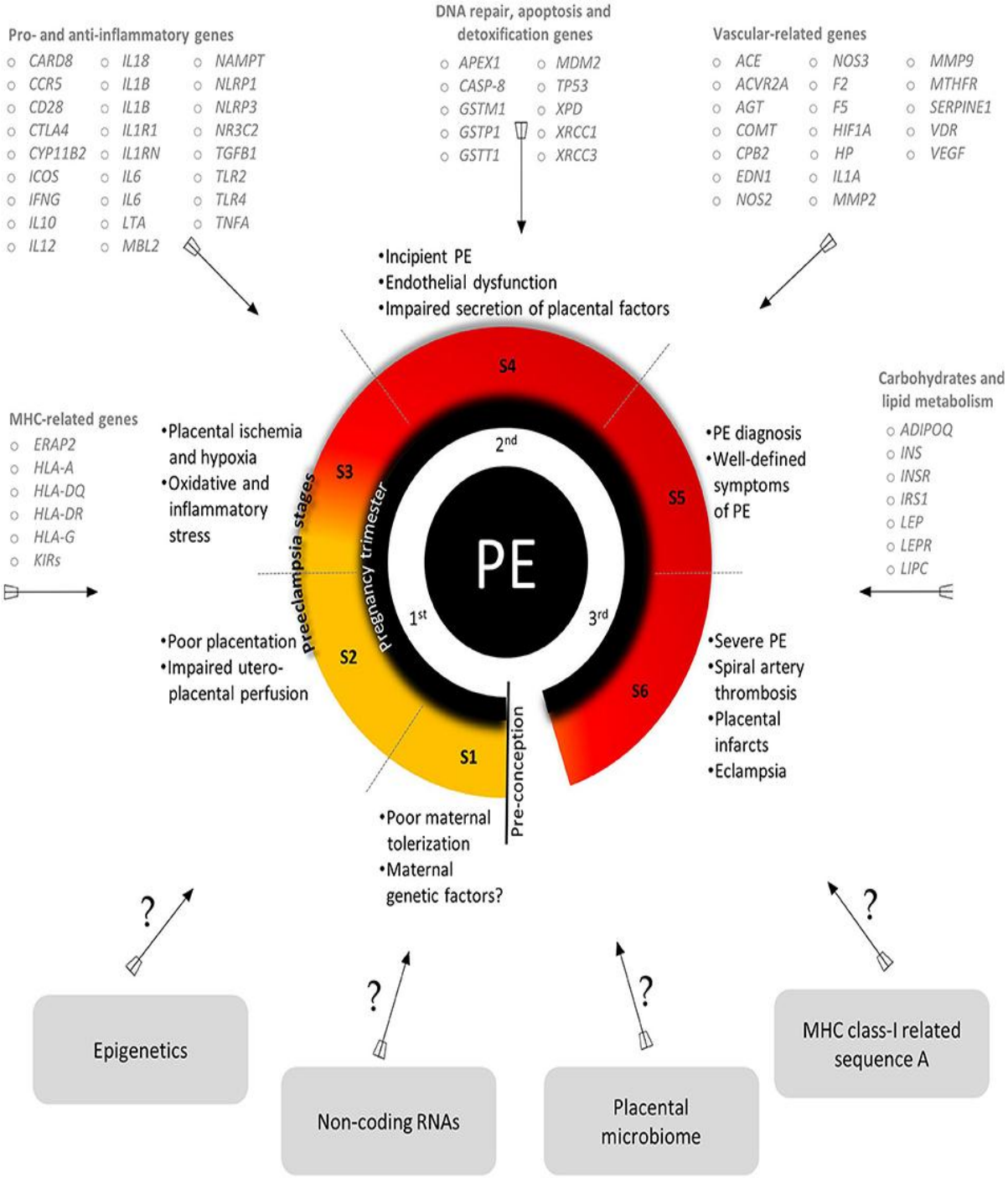


FIGURE 5:

ANATOMY OF UTERINE AND PLACENTAL VASCULATURE

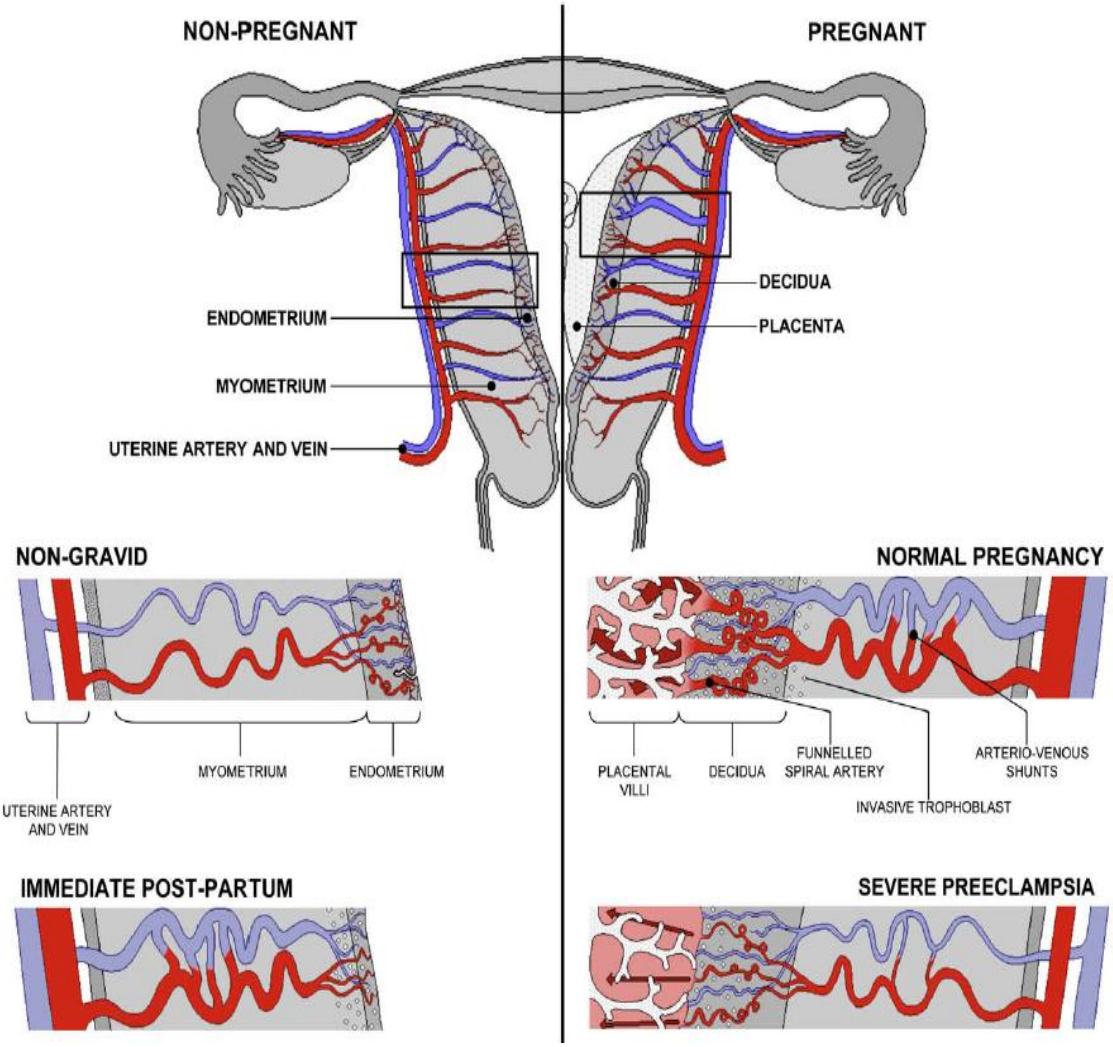


FIGURE 6:

COMPLETE CYTOTROPHOBLAST INVASION IN NORMAL PREGNANCY.

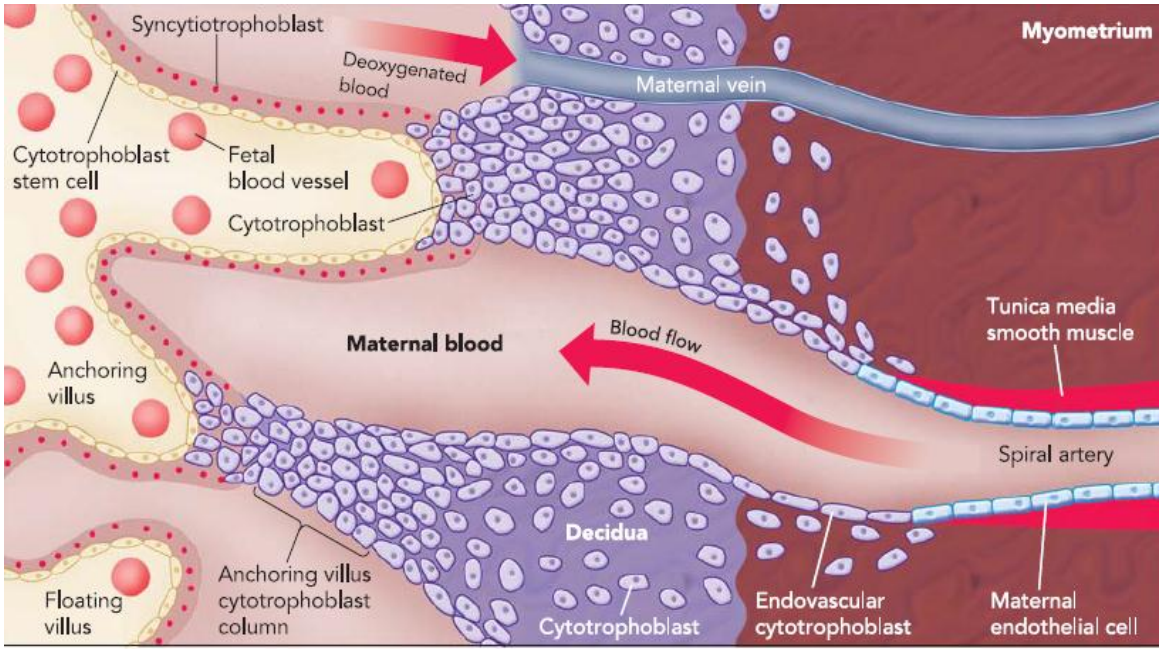


FIGURE 7:

SHALLOW CYTOTROPHOBLAST INVASION IN PRE ECLAMPSIA:[(10)]

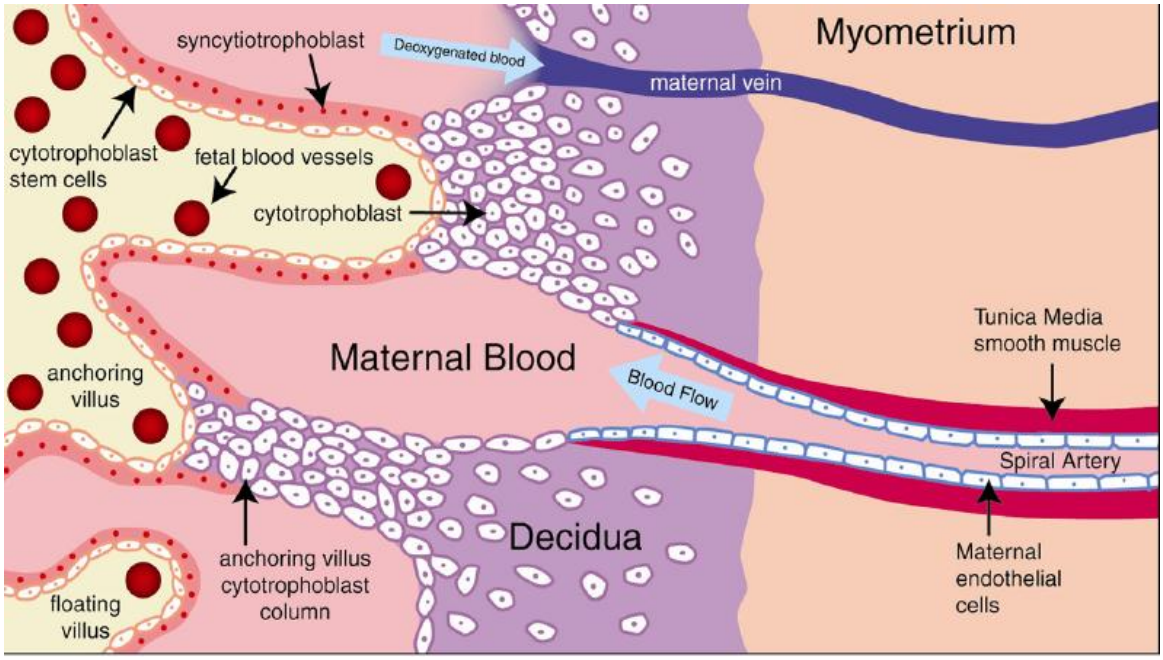


FIGURE 8: ANGIOGENESIS AND ANTIANGIOGENESIS FACTORS IN NORMAL PREGNANCY

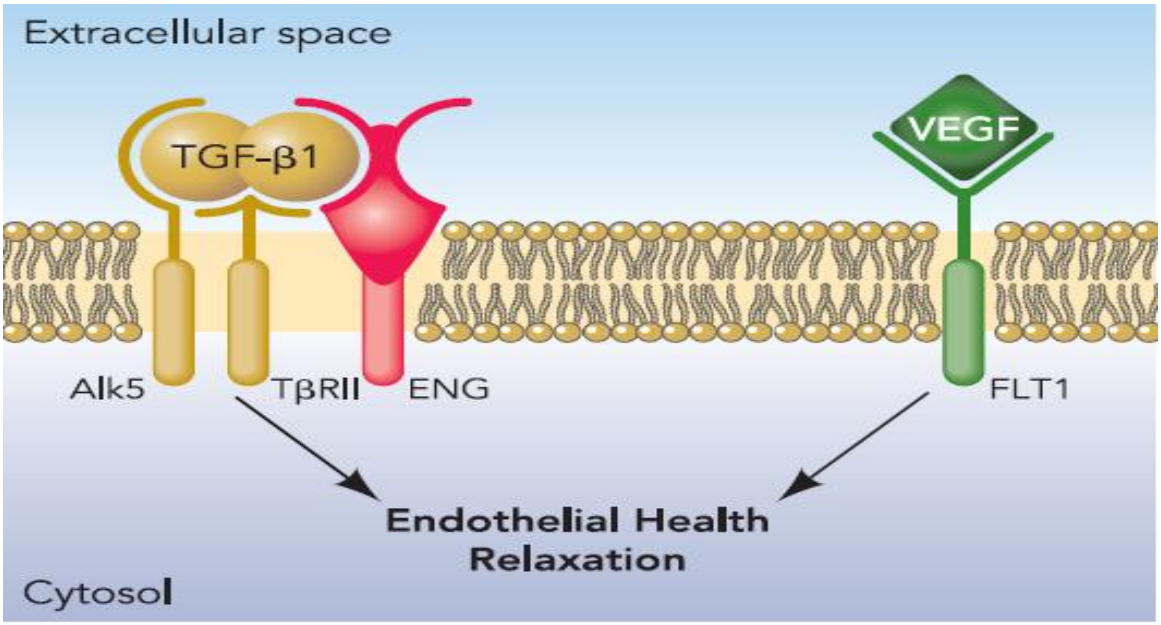


FIGURE 9:

ANGIOGENESIS AND ANTIANGIOGENESIS FACTORS IN PRE ECLAMPSIA.

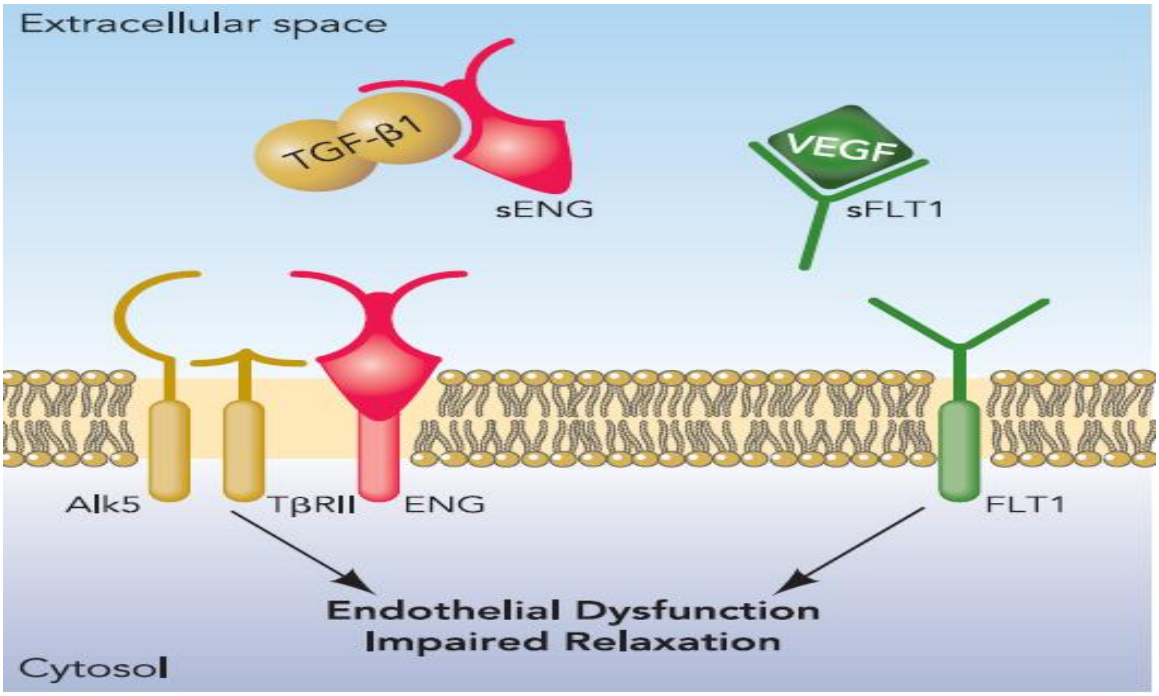


FIGURE 10:

OXIDATIVE STRESS IN PRE ECLAMPSIA

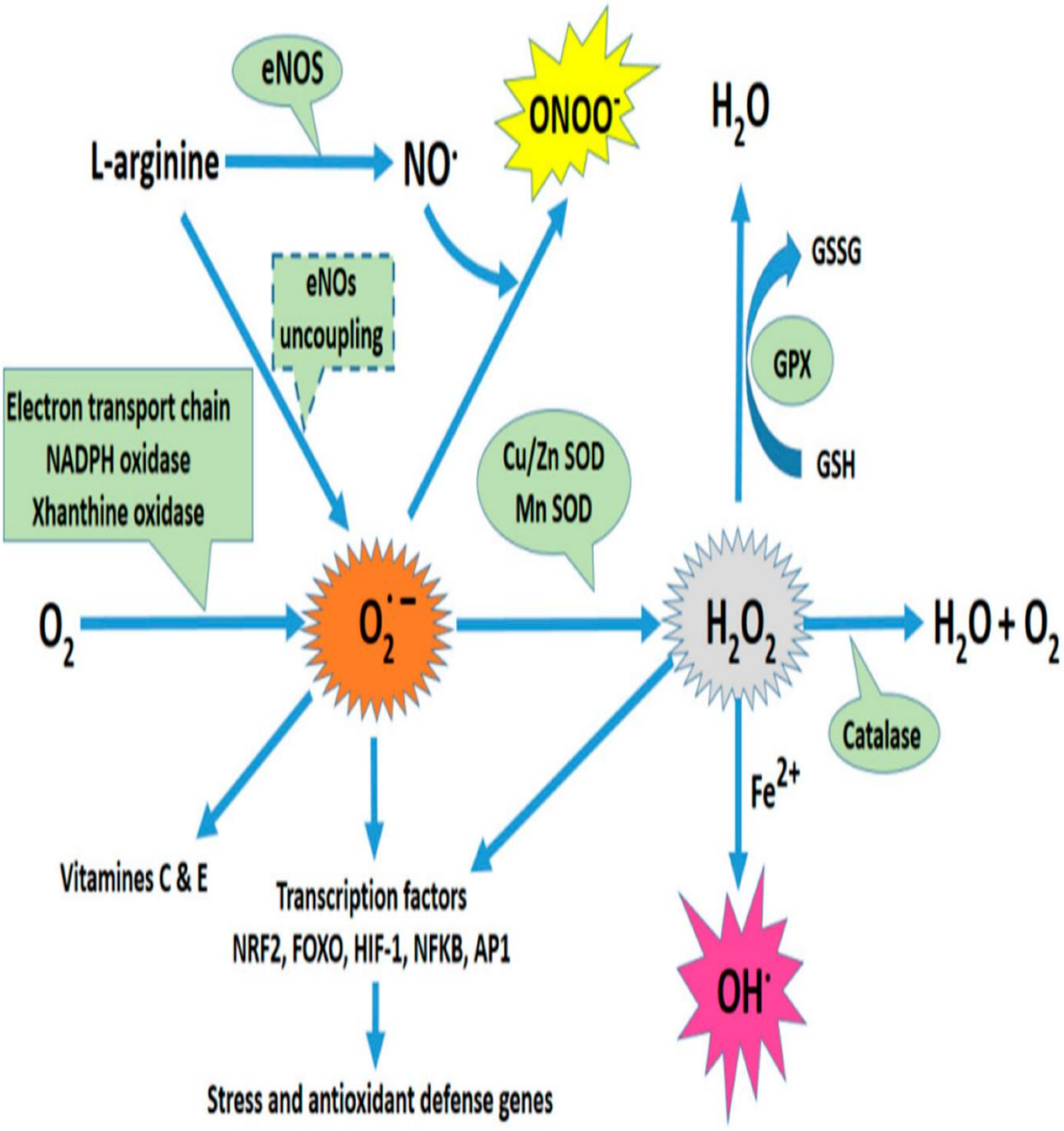


FIGURE 11:

ABSENCE OF Th2 POLARIZATION IN PRE ECLAMPSIA.

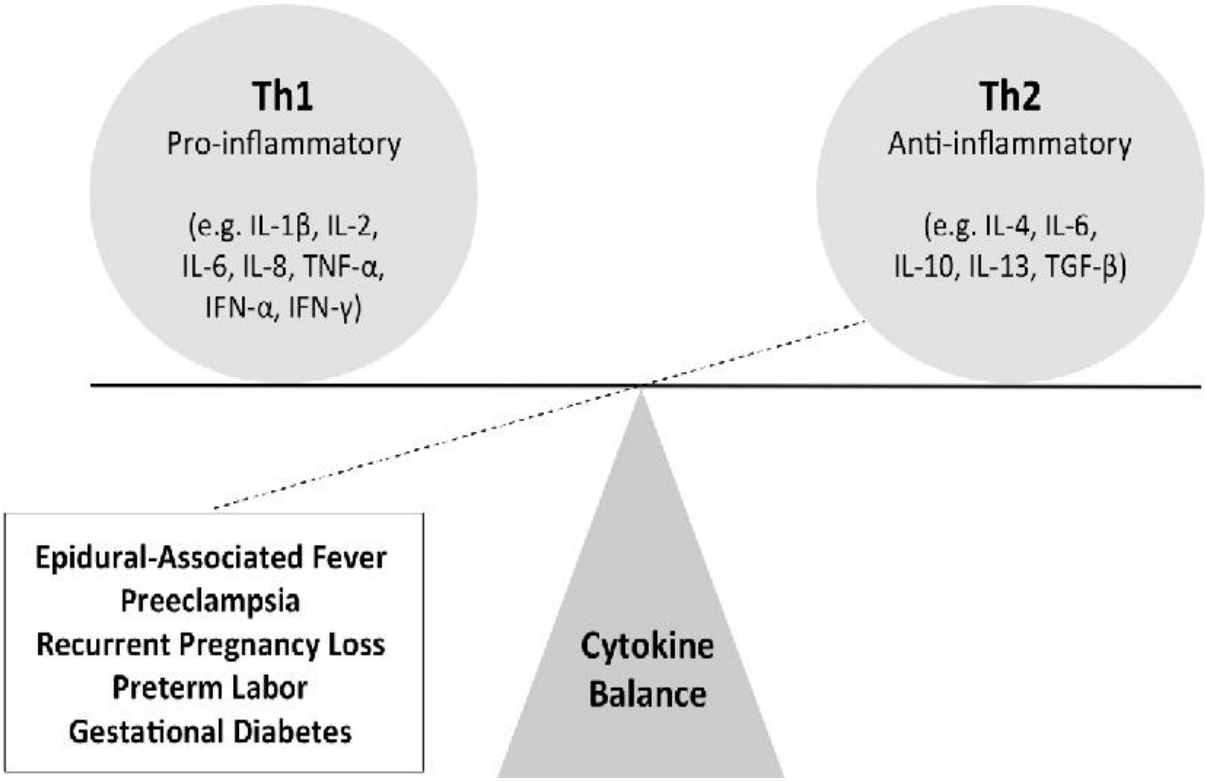


FIGURE 12:

IMMUNOLOGICAL EVENTS IN PRE ECLAMPSIA.

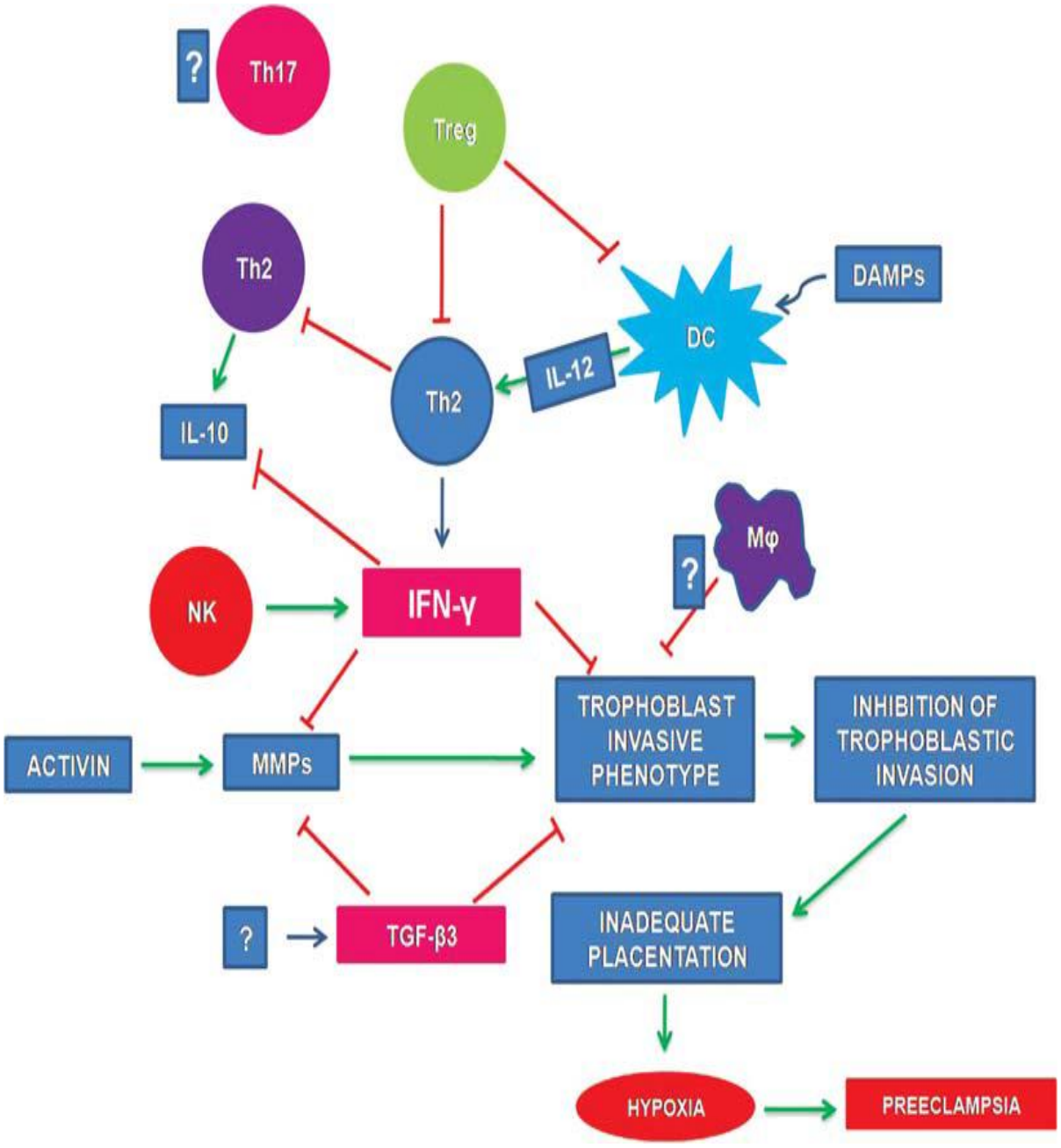


FIGURE 13:

ANGIOTENSIN II RECEPTOR MORPOHOLOGY.

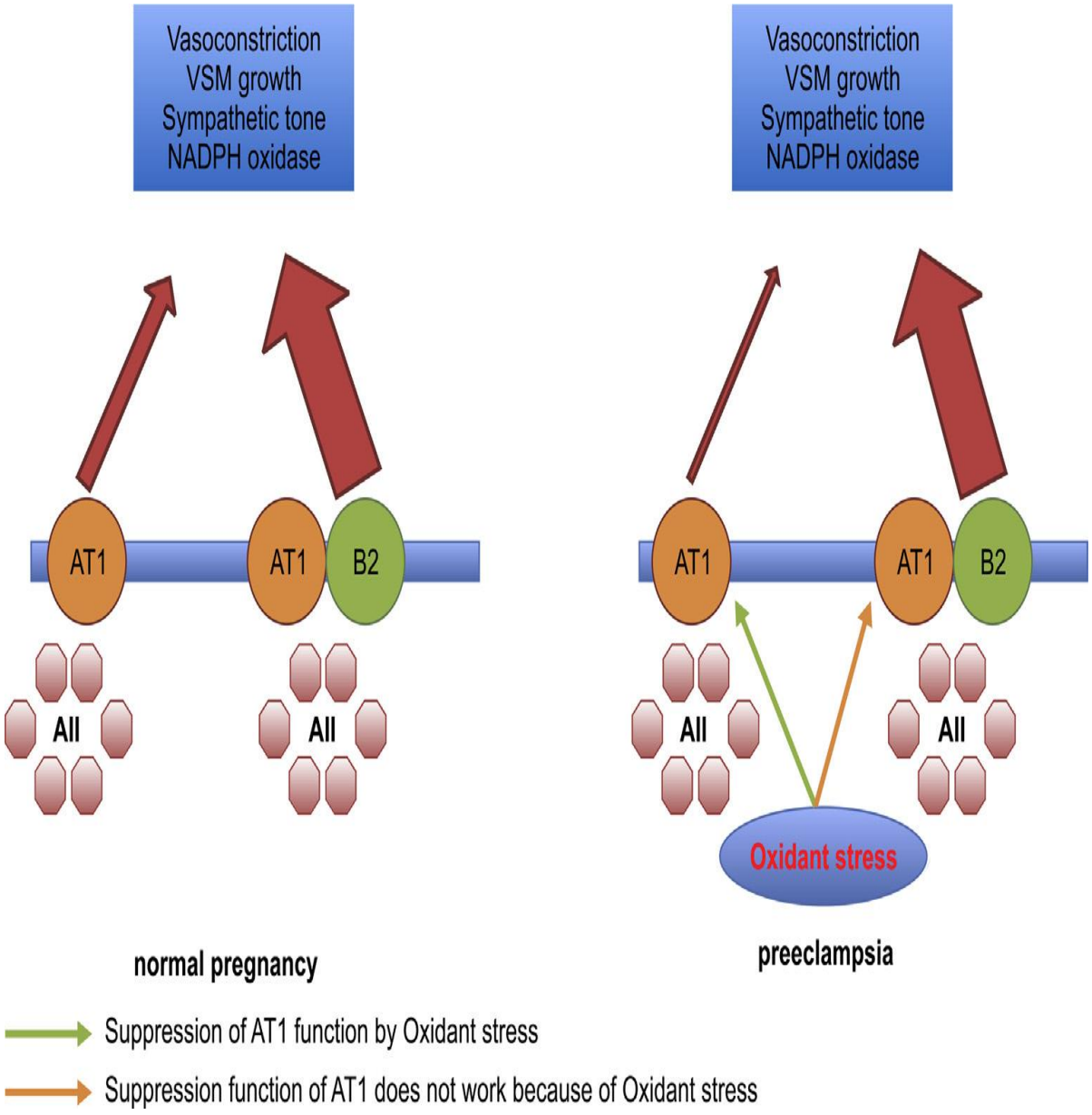


FIGURE 14:

COMPREHESIVE MODEL EFFECTS OF GALECTIN(PP13) IN MATERNAL VASCULATURE.

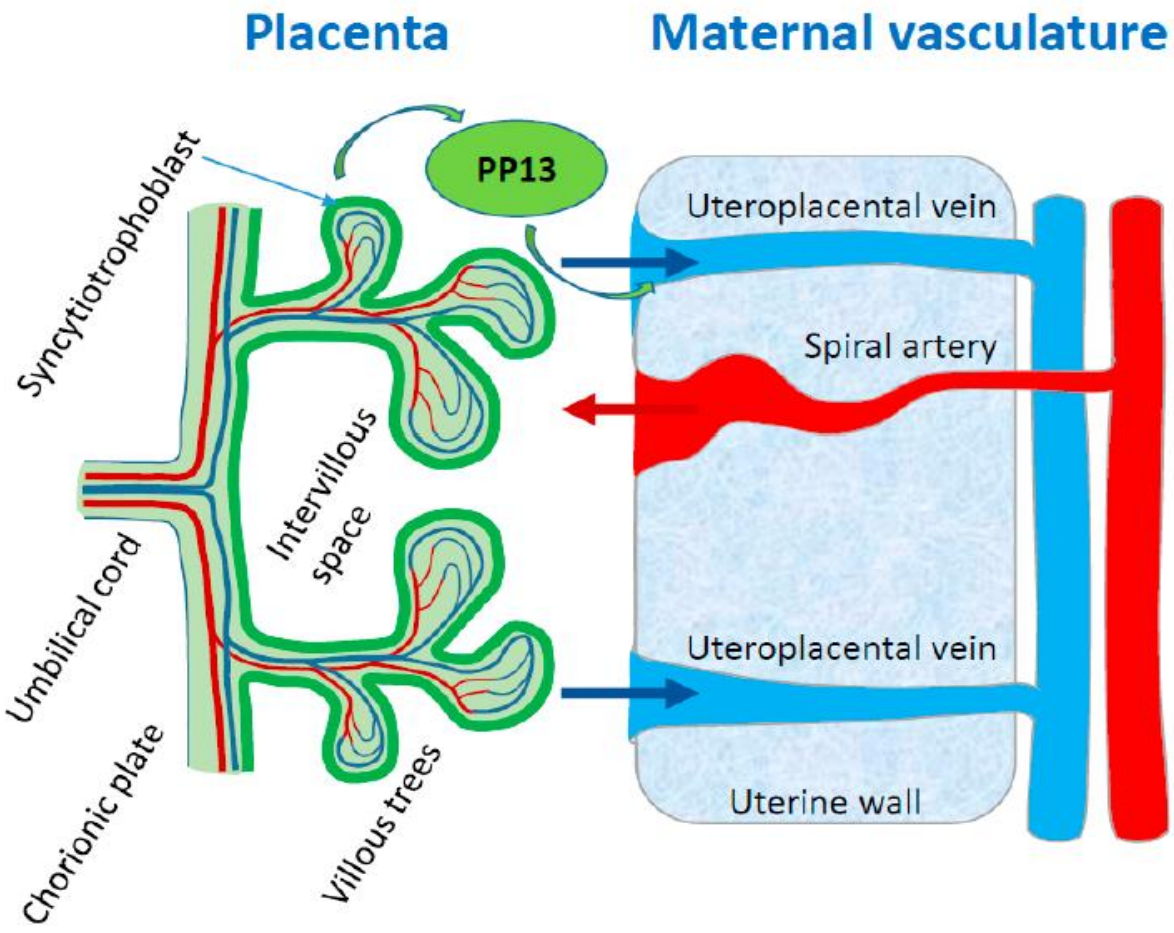


FIGURE 15:

SUMMARY OF ETIOPATHOGENESIS RELATED TO PRE ECLAMPSIA:

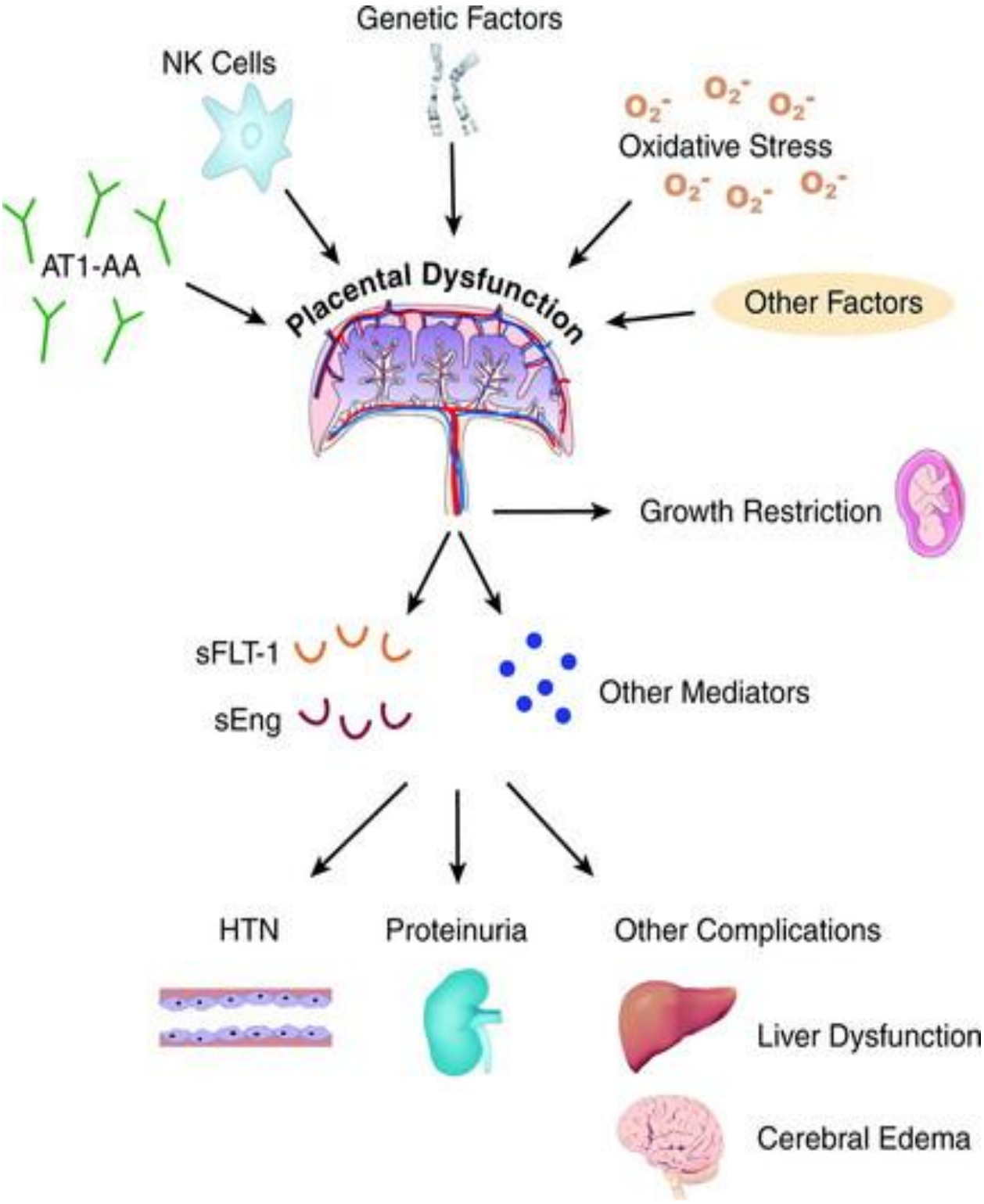


FIGURE 16:

PICTURE ILLUSTRATING SIGNS AND SYMPTOMS IN PRE ECLAMPSIA.

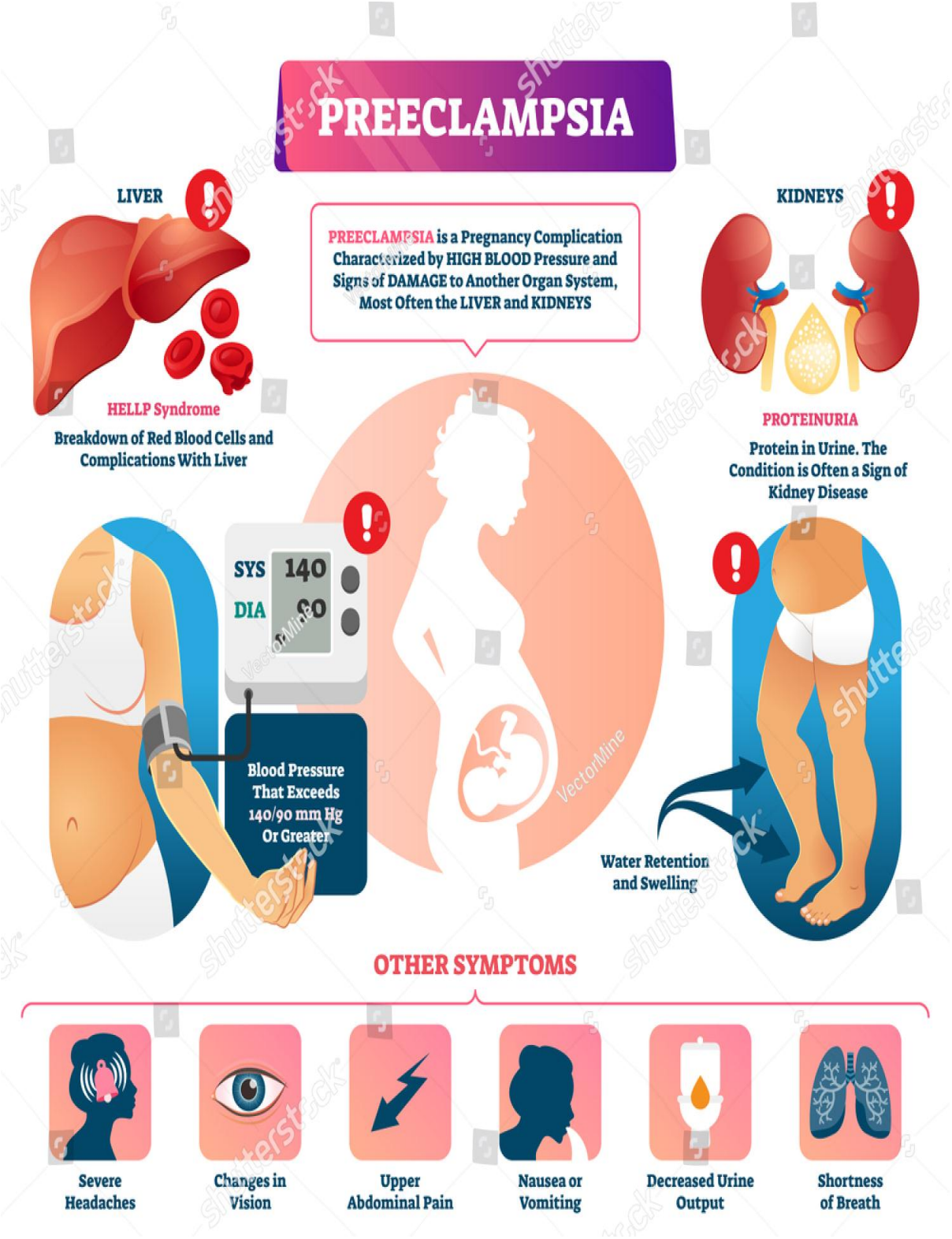
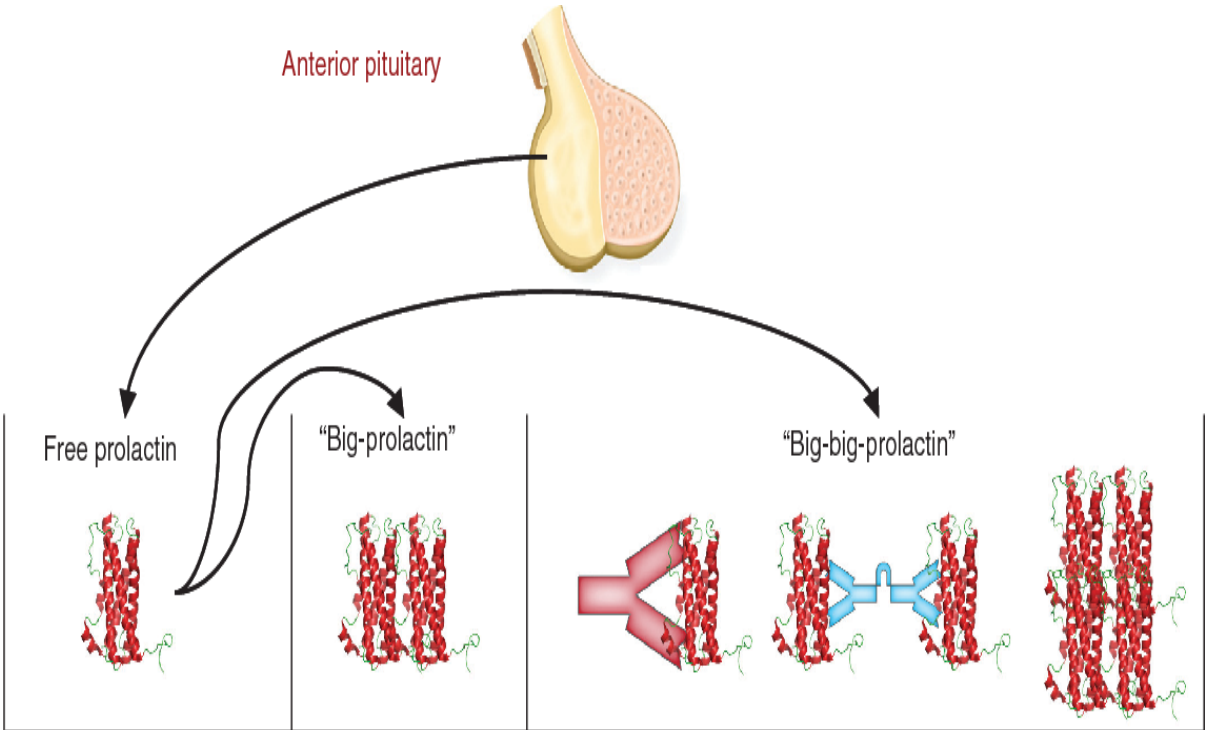


FIGURE 17:

ILLUSTRATES STRUCTURE OF MOLECULAR FORMS OF PROLACTIN[22]



Monomeric	Dimeric	IgG-prolactin	IgA-prolactin	Prolactin aggregates
23 kDa	40–60 kDa	>100–150 kDa		
Active	Inactive	Inactive		
60%–90%	15%–30%	0%–8%	0%–2%	0%–3%

FIGURE 18:

PROLACTIN-SHORT LOOP NEGATIVE FEEDBACK REGULATION :[25]

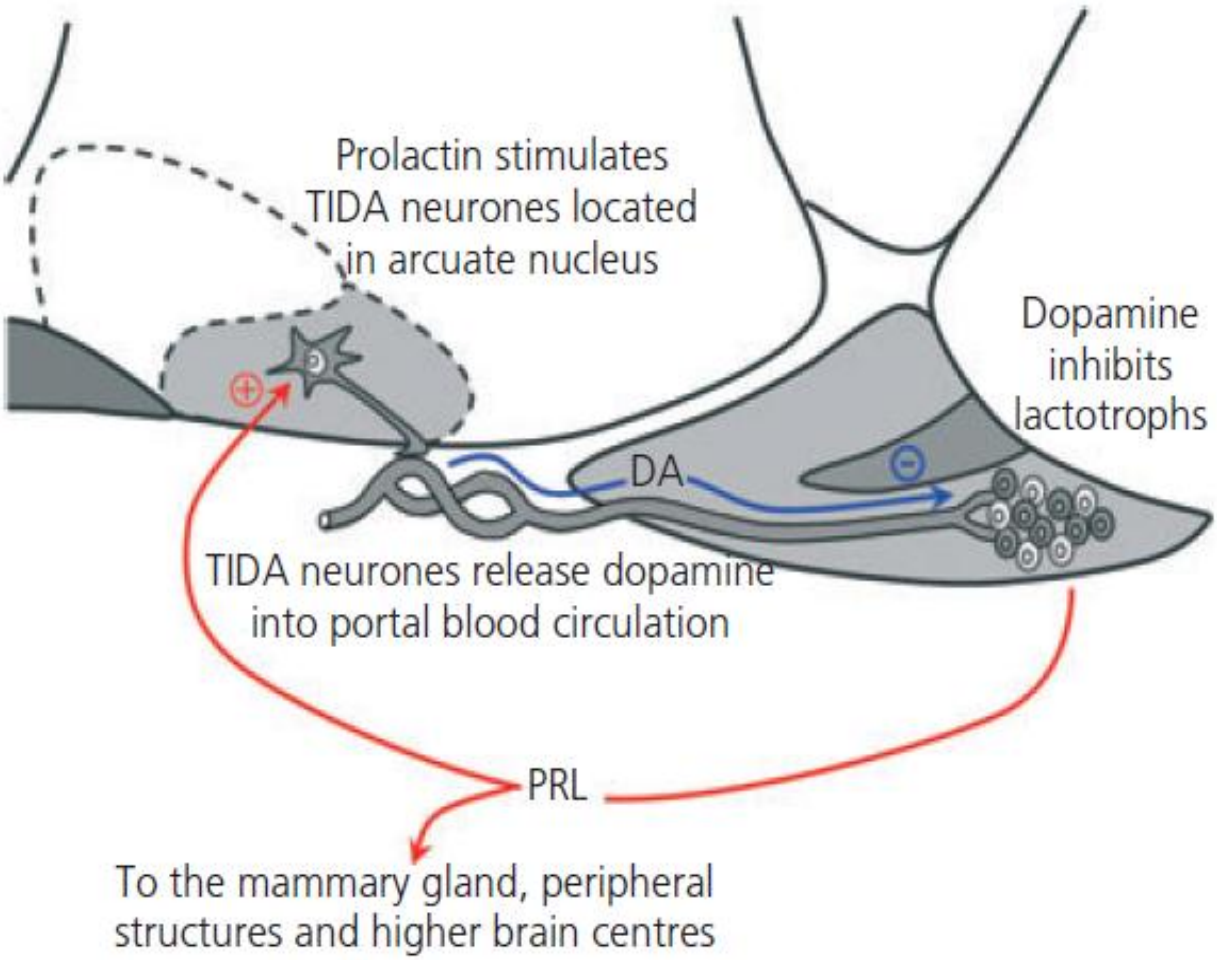


FIGURE 19: PROLACTIN – SIGNAL TRANSDUCTION PATHWAY.[26]

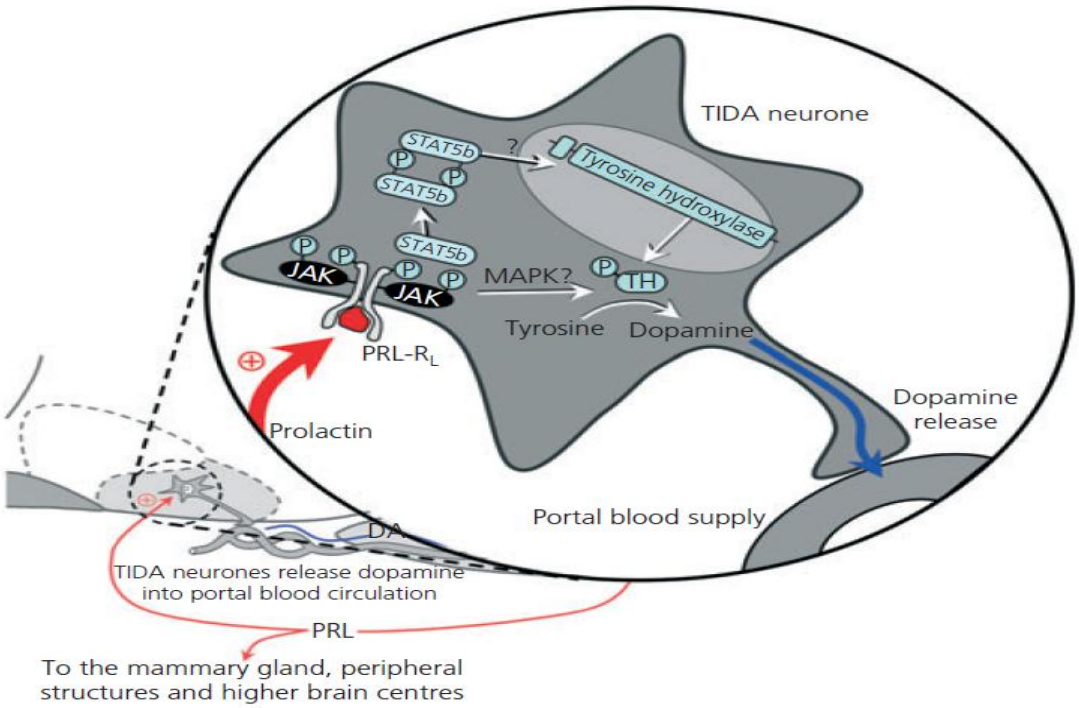


FIGURE 20: INCREASED PROLACTIN IN LATE PREGNANCY.

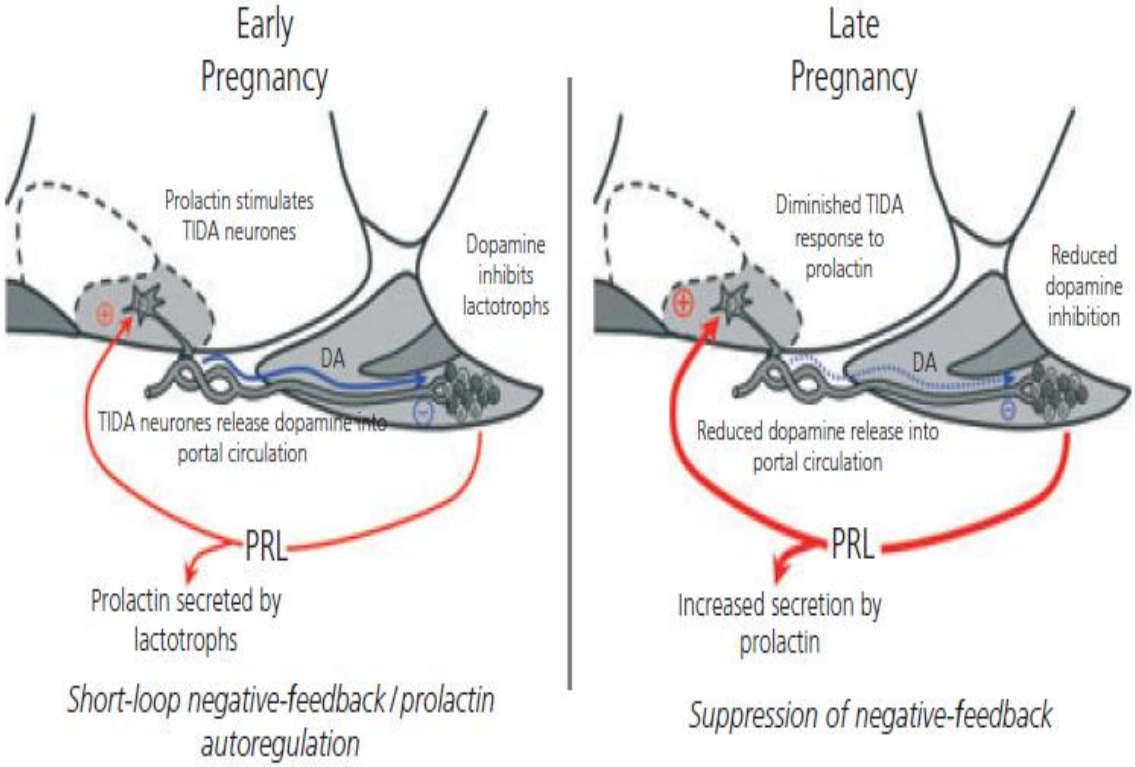


FIGURE 21: ROLE OF PROLACTIN FRAGMENTS IN PRE ECLAMPSIA[21]