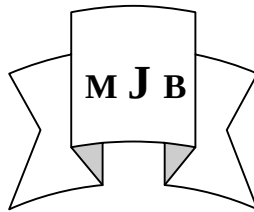


The Relationship between Testosterone Hormone and Lipid Profile, Proteins and Some Trace Elements in the Sera of Patients with Preeclampsia

Ameera Jasem Mohamad Amen Bushra AL-Robaee* Tariq Hufdhay AL-Khayat*

*College of Medicine, University of Babylon, Hilla, Iraq.



Abstract

Preeclampsia is one of the most common diseases which occurs during the second and third trimester of pregnancy. The incidence of this disease is 2-5% among pregnant women.

The aetiology is still in debate and many theories were introduced in this field by many investigators in different countries. It is sometimes called disease of theories due to the contradictory issues concerning its causes and consequences.

In this study we tried to elucidate the relationship between testosterone and some biochemical constituents which vary during pregnancy (i.e , lipid profile, total protein, albumin and minerals (Ca & Mg).

This work was carried out on fifty five pregnant women referred to Babylon Hospital for Obstetric & Paediatrics for the period from November 2007 to May 2008 .The serum samples obtained from those patients and control groups (55 healthy pregnant) were analyzed for lipids, protein and minerals in addition to testosterone .The study group was subdivided into four subgroups as follows :

- 1.Group I is comprised of 25 preeclamptic patients in the second trimester of pregnancy .
2. Group II is comprised of 30 preeclamptic patients in the third trimester of pregnancy .
- 3.Group III includes 25 healthy pregnant women (2nd trimester) which served as control group .
- 4.Group IV represents 30 healthy pregnant subject in their third trimester of pregnancy

The results revealed a significant increase in serum testosterone levels in Group I and Group II compared with Group III and IV ($p<0.01$). These were insignificant decrease in hormone level in Group IV in comparison with Group III ($p=0.36$).

The results showed a significant increase in serum level of total cholesterol, TG, LDL, VLDL in Group I and Group II compared with those in Group III and Group IV, ($p<0.001$), ($p<0.01$) at respectively. However, there was a concomitant decrease in serum HDL level in Group I and Group II when compared with Group III and Group IV.

The results showed also a significant decrease in the levels of total protein ,albumin ,Ca and Mg in preeclamptic women compared with normotensive pregnant ($p<0.05$) .These changes were insignificant when the results of these component in Group IV were compared with Group III ($p>0.05$).

There were significant correlation between serum testosterone levels and lipid profile , protein and minerals .This gives a preliminary idea about the role of testosterone in such changes. There were a positive correlation between testosterone and lipid profile except HDL-C which decrease at increase the testosterone in G1,G2 and G3 ($p<0.01$) and there were positive correlation between cholesterol / albumin ratio and testosterone in G1,G2 and G3 ($p<0.01$) but a negative correlation in G4 ($p<0.01$). There were inverse relationship between cholesterol and albumin ($p<0.001$).

الخلاصة

مرض قبل الشنج هو واحد من الأمراض الأكثر شيوعاً يحدث أثناء الحمل في فصليه الثاني والثالث ونسبة معدل وقوع هذا المرض هي 2-5% بين النساء الحوامل و أسباب هذا المرض غامضة لحد الآن و العديد من النظريات طرحت في هذا المجال من قبل بعض الباحثين وفي مختلف الدول .في بعض الأحيان يدعى هذا المرض بمرض النظريات بسبب تناقض الإصدارات المتعلقة بأسبابه ونتائجه .

و في هذه الدراسة حاولنا ولأول مرة توضيح العلاقة بين هرمون الذكورة وبعض مكونات الكيمياء الحيوية التي تختلف أثناء الحمل (نطاق الدهون , البروتين الكلي , الألبومين , الكالسيوم , المغنيسيوم) .

وقد انتقينا 55 امرأة مريضة حامل أحييت إلى مستشفى بابل للولادة والاطفال للفترة بين كانون الاول 2007_أيار 2008 لإجراء الدراسة وتم تحليل مصول من أولئك المرضى مع مجموعة السيطرة ال(55 امرأة سليمة حامل) وفحصت الدهون والبروتينات وبعض المعادن بالإضافة إلى هرمون الذكورة .

قسمت الدراسة إلى أربع مجاميع فرعية كما يأتي:

- 1- المجموعة الأولى: تشمل 25 مريضة مصابة بمرض قبل الشنج في الفصل الثاني من الحمل .
 - 2- المجموعة الثانية: تشمل 30 مريضة مصابة بمرض قبل الشنج في الفصل الثالث من الحمل .
 - 3- المجموعة الثالثة: تشمل 25 امرأة سليمة حامل في الفصل الثاني من الحمل.
 - 4- المجموعة الرابعة: تشمل 30 امرأة سليمة حامل في الفصل الثالث من الحمل.
- وقد أظهرت النتائج زيادة معنوية في مستوى هرمون الذكورة في المجموعة الأولى والثانية مقارنة بالمجموعة الثالثة والرابعة ($p<0.001$) وهناك نقصان غير معنوي في هرمون الذكورة في المجموعة الرابعة مقارنة بالمجموعة الثالثة ($p=0.36$). وبينت النتائج إن هناك زيادة معنوية في الكوليسترول الكلي وتراي كليسايد وكوليسترول الليبوبروتين عالي الكثافة وكوليسترول الليبوبروتين قليل الكثافة جداً في المجموعة الأولى والثانية بالمقارنة مع المجموعة الثالثة والرابعة ($p<0.001$), ($p<0.01$) بالتعاقب بينما هناك نقصان معنوي في كوليسترول الليبوبروتين عالي الكثافة في المجموعة الأولى والثانية عند المقارنة بالمجموعة الثالثة والرابعة .

كما أظهرت النتائج نقصاناً معنوياً في مستوى البروتين الكلي والألبومين والكالسيوم والمغنيسيوم في النساء المصابات بمرض قبل الشنج بالمقارنة مع الحوامل ذوات الضغط الطبيعي ($p<0.05$) وهذه التغيرات كانت غير معنوية في المجموعة الثالثة والرابعة ($p>0.05$).

وقد أظهرت النتائج وجود علاقة معنوية بين مستوى هرمون الذكورة في المصل و(نطاق الدهون والبروتين والمعادن). وهذا يعطي فكرة حول دور هرمون الذكورة في هذه التغيرات. وتوجد علاقة موجبة بين هرمون الذكورة ونطاق الدهون ($p<0.01$) باستثناء كوليسترول الليبوبروتين عالي الكثافة الذي يقل بزيادة هرمون الذكورة في المجموعة الأولى والثانية والثالثة. وهناك علاقة موجبة بين هرمون الذكورة ونسبة الكوليسترول/الألبومين ($p<0.01$) في المجموعة الأولى والثانية والثالثة ولكن العلاقة موجبة في المجموعة الرابعة وبزيادة الكوليسترول يقل الألبومين ($p<0.001$).

Introduction

Hypertension in pregnancy is a significant problem , if it is associated with proteinuria (which indicates multisystemic disease, known as preeclampsia(PET)), it will be associated with increased morbidity and mortality for both mother and fetus. It is a common problem accounting one from five women after 20 weeks of gestation [1].

Preeclampsia is divided according to severity into mild, moderate and severe forms depending on the level of the blood pressure and the degree of proteinuria , mild preeclampsia characterized by diastolic blood pressure of 90 mmHg with proteinuria less than 5gm/24hr (+) to (++) and edema in feet . In severe preeclampsia , blood pressure is more than 110 mmHg and proteinuria more than 5gm/24hr (+++) to(+++++) and edema in hands and or face [2]. Symptoms and signs include sudden rise in blood pressure , severe proteinuria , generalized edema, excessive weight gain , visual changes such as blurred or double vision , headache , nausea , vomiting , epigastric pain , oliguria, changes in liver or kidney function tests. These are signs and symptoms of imminent eclampsia. If these symptoms are associated with seizure, then the condition is called eclampsia .In PET an increase in the resistance of blood vessels may hinder blood flow in many different organs like the liver, kidney, brain, uterus and placenta affecting their function or causing placental abruption which is a premature separating of the placenta after 20 weeks of gestation . PET can also lead to fetal complications including intrauterine growth restriction (poor fetal growth) and still birth [3].

Major preexisting risk factors for PET include primigravida state , history of PET in previous pregnancy , large body size, a family history of PET, multiple pregnancy , preexisting maternal hypertension, pregestational diabetes , antiphospholipid antibody syndrome , vascular or connective tissue disease and advanced maternal age (> 35 to 40 years) [4].

Preeclampsia was known as the disease of theories, as the exact course of events that leads to the clinical syndrome have not been elucidated. The first theory relates preeclampsia to immunogenic factors. Numerous studies suggest a genetic susceptibility to PET , daughters of women with PET are four to five times more likely to develop the syndrome than daughters in law (5). How the genotype result in the characteristic placental lesion is not known but may involve an immunological defect resulting failure to establish tolerance to the fetal allograft [5,6] .

The second theory relates the syndrome to the disturbance in different vasoactive compounds [6]. Disturbance of endothelial cells in PET leads to alteration in the production of several vasoactive compounds producing a vasoconstrictor state: Prostacyclin (PGI₂) , the predominant vasodilator prostanoid is reduced while placental production of vasoconstrictor thromboxane A₂ is increased. Plasma endothelin ,a potent vasoconstrictor is also increased[5].

The third theory which relates the disease to uteroplacental ischemia, suggests the following:-

- 1- Preeclampsia begins with uteroplacental ischemia , which is an increase intramural resistance in the myometrial vessels, leads to heightened myometrial tension

produced by large fetus in a primipara, twins or hydramnios [6].

2- The uteroplacental ischemia leads to the production of vasoconstrictor substance , which enters the circulation and produces renal vasoconstriction leading to increased production of renin - angiotensin and aldosterone [6].

3-The renin-angiotensin system produces a generalized vasoconstriction and aggravates further the uteroplacental ischemia [6]. It is followed by systemic of cytotoxic products that damage maternal vascular endothelium [7].

4- Aldosterone leads to water and electrolyte retention and generalized edema[8].

Women with cardiovascular (CV) risks are at increased risk for preeclampsia ,and those with history of preeclampsia are at increased risk for post-pregnancy CV morbidity and mortality , compared with women with history of normal pregnancy, This suggests that preeclampsia and CV disease share common pathogenic mechanism. These changes may involve endothelial function deficient in preeclampsia , as seen from reduced prostacyclin and / or elevated endothelin-1 or thromboxane A2 production [9].

Theca cells are the source of androstenedione and testosterone . These are converted by aromatase enzyme in granulosa cell to estrone and estradiol . Significant amounts of estrogens are produced by the peripheral aromatization of androgens[10]. In female , adrenal androgens are important substrates , since as much as 50% of the estradiol E2 produced during pregnancy comes from the aromatization of androgens [10]. Aromatase activity is present in

adipose cells and also in liver, skin and other tissues [11].

Total protein , albumin, globulin and albumin/ globulin (A/G ratio):

The concentration of total protein in human plasma is approximately 6.2 -8.2 gm/dl , and comprises the major part of the solids of the plasma [10]. The major types of protein in the plasma are albumin , globulin and fibrinogen . Albumin constitutes the major part of plasma proteins . It has one polypeptide chain with 585 amino acids and 17 disulfide bonds . It has molecular weight of 69 KD . It is synthesized by hepatocytes . Half life of albumin is about 20 days [12].

A major function of albumin is to provide colloid osmotic pressure in the plasma which prevents plasma loss from the capillaries. Another major function of albumin is to transport various hydrophobic substances. All proteins have buffering capacity and albumin may be considered as the transport form of essential amino acids from liver to extrahepatic cells[12]

The globulins perform a number of enzymatic functions in the plasma , but equally important , they are principally responsible for the body's both natural and acquired immunity against invading organisms [13]. A/G ratio is altered or even reversed by the reticuloendothelial system and decrease in albumin. This again leads to edema [14] . Fibrinogen polymerizes into long fibrin threads during blood coagulation , thereby forming blood clots that help repair leaks in the circulatory system [13].

The total concentration of serum proteins decrease by about 1g/l during pregnancy . Most of the decrease occurs during the first trimester . The decrease is mainly in serum albumin . The maternal antibody (IgG)

component , which is the major immunoglobulin transferred to the fetus , falls progressively, alteration that occurs in the levels of clotting factors and plasminogen is probably brought about by estrogen action on the liver [15].

Mineral homeostasis and hypertension
:

Magnesium ischemia is a term used to denote the functional impairment of the ATP – dependent sodium /potassium and calcium pumps in the cell membranes and within the cell itself .The production of ATP and the functioning of these pumps are magnesium dependent and are critically sensitive to acidosis . Zinc and iron deficiencies may impair these pumps and thus contribute to magnesium ischemia as does acidosis [16] . It refers to functional magnesium deficiency whether actual or induced. It is argued that chronic acidosis is the most common inducing factor . It can also unify clinical thinking about pregnancy – induced hypertension , preeclampsia-eclampsia and acute fatty liver of pregnancy. as well as the coagulopathy of pregnancy . Mg can lead to important predictions about perinatal morbidity and suggests that early supplementation might prevent much pregnancy – induced disease [16] . On the basis of the therapeutic effects of magnesium salts and the knowledge vasodilating properties of magnesium, it was suggested that a deficiency of magnesium contributes to the development of vasoconstriction in preeclampsia [17].

Calcium homeostasis is an important aspect of maternal and fetal physiology during gestation ,and recent evidence implicates alterations in calcium metabolism in the pathogenesis of hypertension during pregnancy. Deficiencies in calcium intake have been linked to preeclampsia-eclampsia , and hypocalciuria and deviations in

both 1,25 (OH)₂ D₃ and PTH have been shown in women with preeclampsia [18].

During the past 7 years , some progress has been made in the prevention of preeclampsia . Specifically , clinical studies have shown that calcium supplementation can significantly reduce the frequency of preeclampsia , especially in populations with a low calcium intake. They have suggested that in such population , calcium supplementation is a safe and effective measure for reducing the incidence of preeclampsia [19], as the levels of free intracellular calcium is a major determinant of vascular smooth muscle tone and consequently vascular resistance [20].

However ,the role of plasma calcium status in normal pregnancy is still discussed contraversally , as well as calcium supplementation in preeclampsia [16]. Although epidemiologic studies have suggested a role for calcium deficiency in the development of preeclampsia , the published information regarding calcium metabolism in preeclampsia is scanty [20] .

Materials and Methods

Patients:

This study was conducted in Babylon Maternity and Pediatrics Teaching Hospital from November 2007 to the end of May 2008. Fifty five pregnant women with preeclampsia (twenty five of them in the second trimester of pregnancy while the rest of them were in the third trimester of pregnancy).

All the patients were nonsmokers, have no other diseases . Detailed history and examination performed. Pregnancy is divided into 1st trimester (1-12 week), 2nd trimester (13-28

week) and 3rd trimester more than 28 weeks. Depending on the gestational age, the patients were divided into two groups:

Preeclampsics in the second trimester G1 :

They were twenty five preeclampsics in the second trimester of pregnancy. Age range 18-37 years (mean age $\pm$ SD=26.29 $\pm$ 5.12 year).Gestational age range 21-28 weeks (mean gestational age $\pm$ SD = 24.14 $\pm$ 3.63 week). Body mass index range = 24.7-50.4 kg/m² (mean body mass index $\pm$ SD = 36.7 $\pm$ 9.87 kg/m²).Systolic blood pressure range 140-170 mmHg (mean Systolic blood pressure $\pm$ SD = 151.4 $\pm$ 10.7mmHg). Diastolic blood pressure range 90-120 mmHg (mean diastolic Blood pressure $\pm$ SD = 98.6 $\pm$ 10.3 mmHg). Mean proteinuria = 100 mg/dl.

Preeclampsics in the third trimester G2:

They were thirty preeclampsics in the third trimester of pregnancy. Age range 18-44 years (mean age $\pm$ SD =24.86 $\pm$ 5.4 year). Gestational age range 29-38 weeks (mean Gestational age $\pm$ SD = 35.57 $\pm$ 3.21 week).Body mass index range 32.1-61.2 kg/m² (mean body mass index $\pm$ SD =46.6 $\pm$ 1.6 kg/m²).Systolic blood pressure range 140-200 mmHg (mean Systolic blood pressure $\pm$ SD = 157.1-13.8 mmHg). Diastolic blood pressure range 90-130 mmHg (mean diastolic Blood pressure $\pm$ SD = 101.4 $\pm$ 10.3 mmHg). Mean proteinuria = 300 mg/dl.

Control:

Fifty five apparently healthy pregnant women (twenty five of them were in the second trimester and thirty of them were in the third trimester). Pregnant women with chronic medical problems were excluded from this

study. Depending on the gestational age, the pregnant women were divided into two groups:

Control pregnant women in the second trimester G3:

They were twenty five healthy (normotensive) women in the second trimester of pregnancy. Age range 19-35 years (mean age $\pm$ SD = 23.73 $\pm$ 3.73 year). Gestational age range 20-28 weeks (mean $\pm$ SD =23.43 $\pm$ 3.2 week).Body mass index range 21.4-50.4 kg/m² (mean body mass index $\pm$ SD =35.8 $\pm$ 11.7 kg/m²).Systolic blood pressure range 100-130 mmHg (mean Systolic blood pressure $\pm$ SD = 105 $\pm$ 15.1 mmHg). Diastolic blood pressure range 55-75 mmHg (mean diastolic Blood pressure $\pm$ SD = 64.3 $\pm$ 7.9 mmHg). Proteinuria range <30 mg/dl.

Control pregnant women in the third trimester G4:

They were thirty healthy (normotensive) women in the third trimester of pregnancy. Age range 19-42 year (mean age $\pm$ SD = 26.6 $\pm$ 3.74 year). Gestational age range 29-40 weeks (mean $\pm$ SD = 35.3 $\pm$ 3.99 week). Body mass index range 29.5-45.1 kg/m² (mean body mass index $\pm$ SD = 37.7 $\pm$ 5.9 kg/m²). Systolic blood pressure range 90-130 mmHg (mean systolic blood pressure $\pm$ SD = 110-14.1 mmHg). Diastolic blood pressure range 50-80 mmHg (mean diastolic Blood pressure $\pm$ SD = 62.9 $\pm$ 9.9 mmHg). Proteinuria range <30 mg/dl

Methods :

Serum testosterone,total serum cholesterol,HDL,triglycerides were measured by colorimetric assay . also serum total protein ,albumin,globulin,S.calcium,S.magnisi um all measured by colorimetric assay.

Calculation of body mass index(21):

Body mass index (BMI)
calculated as weight (kg)/height(m)² ,
normal value 18.5 - 24.9 kg/m² .

Statistical analysis:

The statistical analysis is based on ANOVA test to determine the differences between groups and within groups. Correlation, regression and correlation coefficient (r) , using SPSS (statistical product and service solutions) program for data

Results

Testosterone:

Serum testosterone was significantly higher in preeclamptic groups(G1&G2) compared with normal pregnant women groups (G3&G4).Also serum testosterone was significantly higher in G2 compared with G1,and also shows nonsignificant decrease in G4 compared with G3 ,[Fig(1),Table(1),(2)]

Table 1 Serum data of testosterone in preeclamptic and normal pregnant women (2nd and 3rd trimester) (mean $\pm$ SD)

Measured parameter	G1	G2	G3	G4
Testosterone(ng/ml) $\pm$ SD	1.46 $\pm$ 0.199	2.41 $\pm$ 0.54	0.82 $\pm$ 0.198	0.74 $\pm$ 0.24

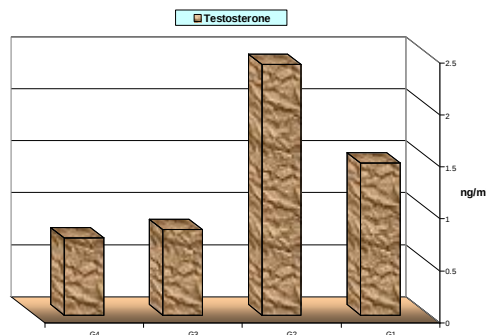


Figure 1 Serum data of testosterone in preeclamptic and normal pregnant women (2nd and 3rd trimester)

Table 2 Significance value for testosterone in different groups

Groups	P value
G1 vs G2	<0.001
G1 vs G3	<0.01
G1 vs G4	<0.001
G2 vs G3	<0.01
G2 vs G4	<0.01
G3 vs G4	=0.36

Lipid profile (total cholesterol, TG, LDL-C, VLDL-C & HDL-C):

Serum total cholesterol, TG, LDL-C and VLDL-C were significantly higher in preeclamptic groups (G1&G2) compared with

normal pregnant groups (G3&G4). This parameters were significantly higher in G2 compared with G1 and in G4 compared with G3, but serum HDL-C was significantly lower in G2 compared with G1 and G4 compared with G3 [Fig (2), Table (3), (4)]

Table 3 Serum data of total cholesterol, HDL-C, TG, VLDL-C, LDL-C in preeclamptic and normal pregnant women (2nd and 3rd trimester) (mean $\pm$ SD)

Measured parameter	G1	G2	G3	G4
Cholesterol (mmol/l)	5.06 $\pm$ 0.167	5.9 $\pm$ 0.292	4.46 $\pm$ 0.71	5.19 $\pm$ 0.82
HDL-C (mmol/l)	1.226 $\pm$ 0.061	1.05 $\pm$ 0.166	1.57 $\pm$ 0.116	1.21 $\pm$ 0.29
TG (mmol/l)	1.59 $\pm$ 0.117	2.72 $\pm$ 0.54	1.28 $\pm$ 0.39	1.89 $\pm$ 0.68
VLDL-C (mmol/l)	0.72 $\pm$ 0.053	1.24 $\pm$ 0.24	0.58 $\pm$ 0.17	0.86 $\pm$ 0.31
LDL-C (mmol/l)	3.117 $\pm$ 0.18	3.62 $\pm$ 0.24	2.31 $\pm$ 0.65	3.12 $\pm$ 0.82

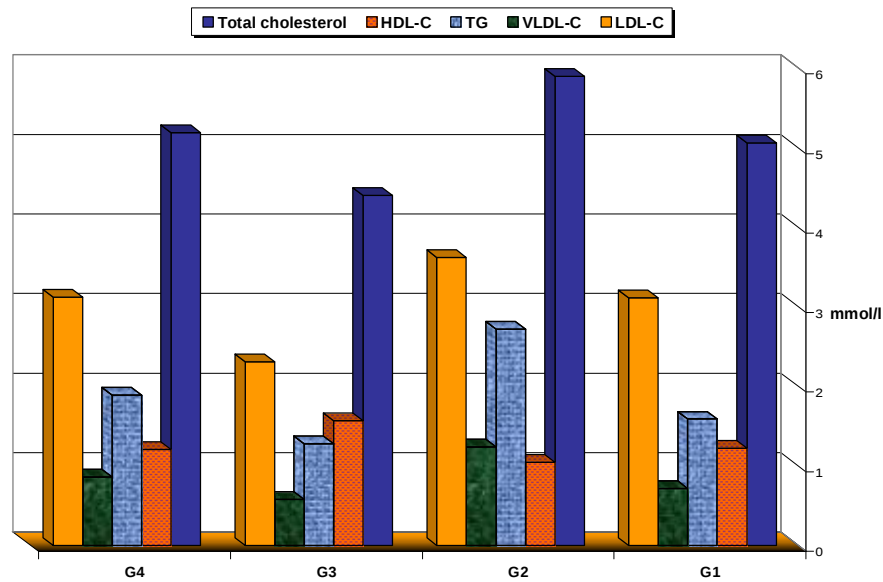


Figure 2 Serum data of total cholesterol, HDL-C, TG, VLDL-C, LDL-C in preeclamptic and normal pregnant women (2nd and 3rd trimester)

Table 4 Significance value for lipid profile in different groups

Groups	P value
G1 vs G2	<0.05
G1 vs G3	<0.001
G1 vs G4	>0.05
G2 vs G3	<0.05
G2 vs G4	<0.001
G3 vs G4	<0.05

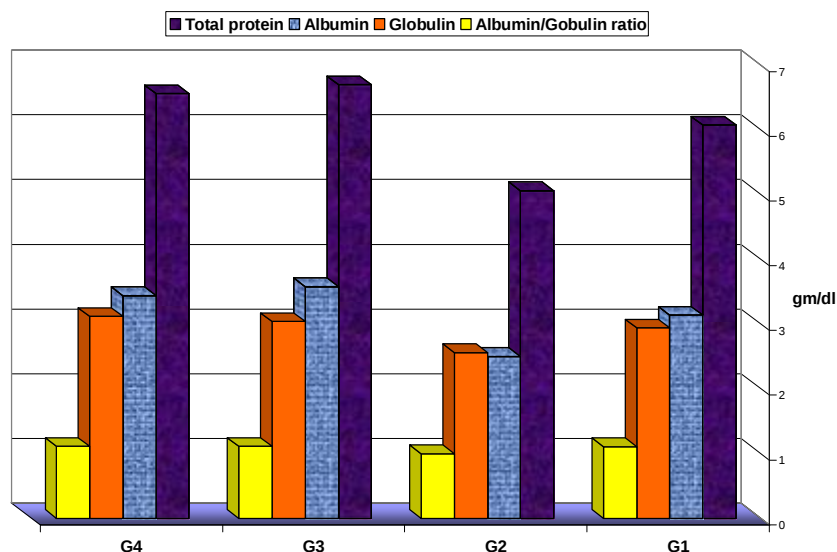
Total protein, Albumin, Globulin and A/G ratio:

Serum total protein and albumin were significantly lower in preeclamptic groups (G1&G2) compared with normal pregnant groups (G3&G4), these

results were significantly lower in G2 than G1 and there was insignificant decrease in G4 compared to G3. The results were reversed for globulin. A/G ratio was significantly lower in G2 than G1 and nonsignificant difference between G3&G4 [Fig(3), Table(5), (6), (7)].

Table 5 Serum total protein, albumin, globulin, albumin/globulin ratio in preeclamptic and normal pregnant women (2nd and 3rd trimester) (mean $\pm$ SD)

Groups	P value
G1 vs G2	<0.001
G1 vs G3	<0.01
G1 vs G4	<0.05
G2 vs G3	<0.01
G2 vs G4	<0.01
G3 vs G4	>0.127

**Figure 3** Serum total protein, albumin, globulin, albumin/globulin ratio in preeclamptic and normal pregnant women (2nd and 3rd trimester) .

Measured parameter	G1	G2	G3	G4
Total protein(gm/dl)	6.076±0.34	5.06±1.22	6.7±0.13	6.56±0.28
Albumin (gm/dl)	3.14 ±0.31	2.5±0.54	3.58±0.12	3.44±0.22
Globulin (gm/dl)	2.94±0.091	2.56±0.69	3.04±0.082	3.12±0.21
A/G ratio	1.109±0.1	0.999±0.11	1.112±0.049	1.112±0.153

Table 6 Significance value for total protein and albumin in different groups

Table 7 Significance value for globulin and albumin/ globulin ratio at different groups

Groups	P value
G1 vs G2	<0.01
G1 vs G3	>0.01
G1 vs G4	>0.05
G2 vs G3	<0.01
G2 vs G4	<0.01
G3 vs G4	<0.01

Total cholesterol/albumin ratio:

Serum total cholesterol/albumin were significantly higher in preeclamptic groups (G1&G2)

compared with normal pregnant groups (G3&G4). This parameter were higher in G2 compared with G1 and in G4 compared with G3.[Fig4),Table8), 9]

Table 8 Total cholesterol/albumin in preeclamptic and normal pregnant women(2nd and 3rd trimester) (mean±SD).

Measured parameter ± SD	G1	G2	G3	G4
Cholesterol/albumin ratio± SD	0.063±0.009	0.096±0.028	0.048±0.009	0.059±0.0122

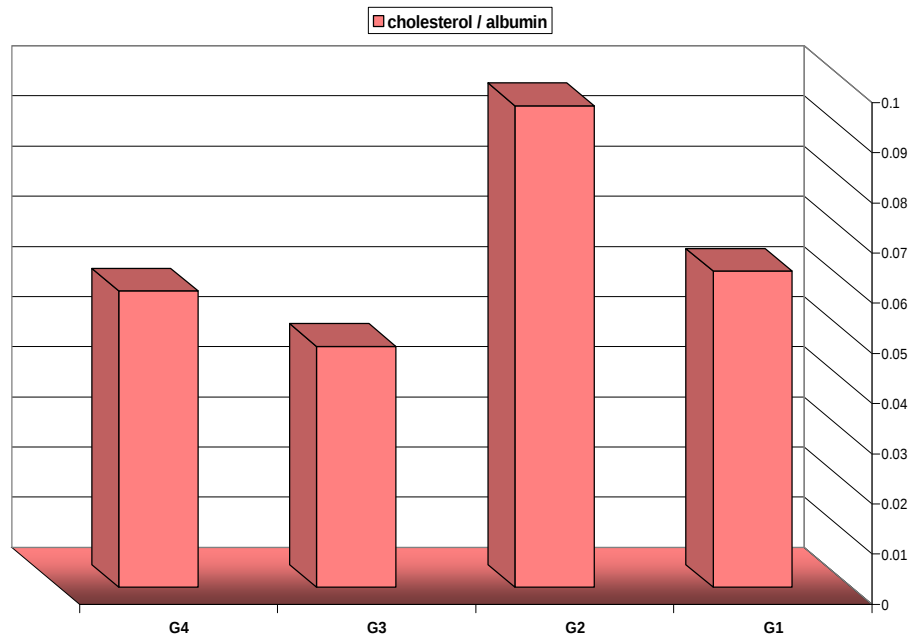


Figure 4 Total cholesterol/albumin in preeclamptic and normal pregnant women (2nd and 3rd trimester)

Table 9 Significance value for total cholesterol/albumin in different groups

Groups	P value
G1 vs G2	<0.001
G1 vs G3	<0.01
G1 vs G4	>0.05
G2 vs G3	<0.001
G2 vs G4	<0.001
G3 vs G4	<0.01

Minerals:

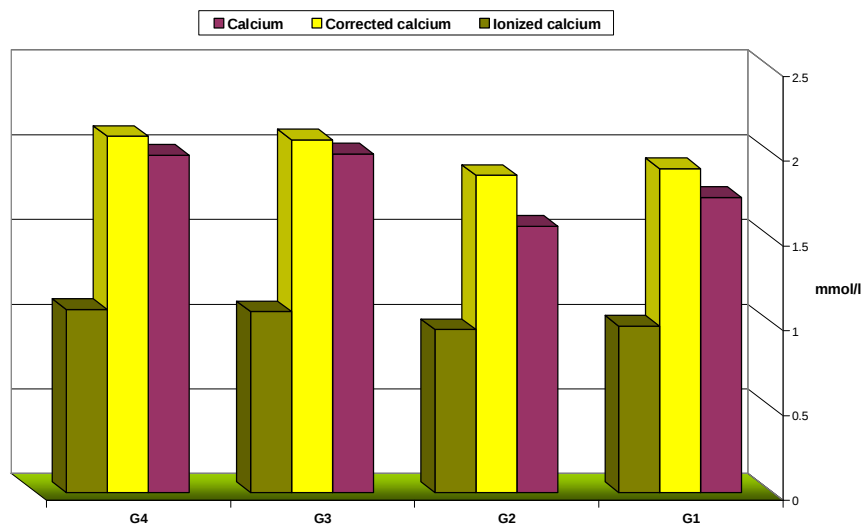
Total calcium, corrected calcium and ionized calcium

Total calcium, corrected calcium and ionized calcium were lower in preeclamptic groups (G1&G2) compared with normotensive groups(G3&G4). These parameters

reversed a significant decrease in G2 in comparison with G4. The results showed insignificant decrease of total calcium in normotensive pregnant at the third trimester in comparison with those of 2nd trimester. Corrected calcium and ionized calcium were higher in G4 than G3, non significant difference [Fig(5), Table(10) , (11) ,(12)].

Table 10 Serum total calcium, corrected calcium, ionized calcium in preeclamptic and normal pregnant women (2nd and 3rd trimester) (mean $\pm$ SD)

Measured parameter	G1	G2	G3	G4
Calcium (mmol/l)	1.74 $\pm$ 0.07	1.57 $\pm$ 0.112	1.997 $\pm$ 0.029	1.99 $\pm$ 0.21
Corrected calcium (mmol/l)	1.91 $\pm$ 0.018	1.87 $\pm$ 0.009	2.08 $\pm$ 0.009	2.101 $\pm$ 0.197
Ionized calcium (mmol/l)	0.98 $\pm$ 0.009	0.96 $\pm$ 0.016	1.066 $\pm$ 0.004	1.079 $\pm$ 0.109

**Figure 5** Serum total calcium, corrected calcium, ionized calcium in preeclamptic and normal pregnant women (2nd and 3rd trimester)**Table 11** Significance value for calcium in different groups

Groups	p Value
G1 vs G2	<0.05
G1 vs G3	<0.01
G1 vs G4	<0.01
G2 vs G3	<0.01
G2 vs G4	<0.001
G3 vs G4	=0.825

Table 12 Significance value for ionized calcium in different groups

Groups	p Value
G1 vs G2	=0.375
G1 vs G3	<0.05
G1 vs G4	<0.01
G2 vs G3	<0.001
G2 vs G4	<0.001
G3 vs G4	=0.386

Total magnesium:

Serum total magnesium was significantly lower in preeclampsia women (G1&G2) compared with

normotensive pregnant women (G3&G4). These results showed nonsignificant difference between G3&G4 and nonsignificant difference between G1&G2[Fig(6),Table(13),(14)].

Table 13 serum magnesium in preeclamptic and normal pregnant women (2nd and 3rd trimester) (mean $\pm$ SD).

Measured parameter	G1	G2	G3	G4
Magnesium (mmol/l)	0.698 $\pm$ 0.029	0.58 $\pm$ 0.08	0.78 $\pm$ 0.064	0.77 $\pm$ 0.23

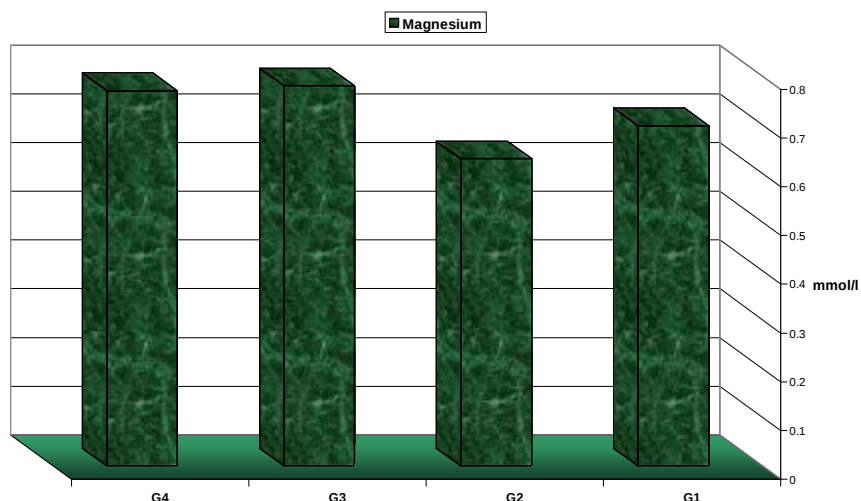
**Figure 6** serum magnesium in preeclamptic and normal pregnant women (2nd and 3rd trimester)

Table14 Significance value for magnesium at different groups

Groups	
G1 vs G2	=0.085
G1 vs G3	=0.024
G1 vs G4	=0.48
G2 vs G3	<0.01
G2 vs G4	<0.001
G3 vs G4	=0.694

Correlation between serum testosterone and other parameters in different groups:

Correlation between serum testosterone and total cholesterol

A significant positive correlations between serum total-cholesterol and testosterone level was noticed in different groups except normal pregnant in third trimester(G4),which reversed negative correlation .Fig(7),(8),(9),(10).

Discussion

Many previous studies reported the changes in oestrogen levels during normal and complicated pregnancy . Besides , there are numerous studies concerning the role of metabolic syndrome in the aetiology of preeclampsia [22,23,24] .

Recent studies revealed the association between the change of serum oestrogen levels and many biochemical parameters during the second and third trimesters of both normal and complicated pregnancy[25]. Other studies connected between those biochemical changes and serum changes of hCG levels in different types of pregnancy[26] .

Depending on the available data, there is no study which link the changes in testosterone levels and consequent changes in lipid profile (as one of the criteria associated with metabolic syndrome).

In this study we tried to elucidate such relationship in order to pave the way for subsequent studies .

We found in our study that BMI are increase in G1& G2 more than G3&G4 ($P<0.001$) but non significant difference between G1 & G3 ($p>0.05$) and significant difference between G2 & G4 ($p<0.001$).

In this study, we found that ten patients out of fifty five preeclamptic patients give positive family history of preeclampsia (18.1%) ; five patients had a previous history of preeclampsia (9%) (both these factors are associated with more incidence of preeclampsia).

In this study , levels of serum testosterone were found to be significantly higher in women with preeclampsia than in normotensive women with similar gestational age . Such increase in hormone level in both 2nd and 3rd trimester can be attributed to :

Low expression of the aromatase gene due to small or impaired for the

conversion of testosterone to estrogen . The decrease of enzyme activity lead to a subsequent increase in testosterone level [24].

In the late pregnancy , when the fetal adrenal gland become mature it will result in further increment in the level of testosterone by conversion of DHEA to testosterone [24].

Human chorionic gonadotropin increase in PET and this will stimulates the ovarian thecal cell to synthesis androstenedione and testosterone [27].

Insulin stimulate the production of testosterone by ovarian tissue which suggests that hyperinsulinemia could be primary change that triggered the increased release of testosterone . However, hyperinsulinemia should also stimulate the production of adrenal androgen [28].

The decrease in testosterone clearance in normal pregnancy is intensified in PET patients. This will lead to increase in serum testosterone levels [29].

Our results were in good agreement with the results reported by Golmahamed -Is [30] and Jasim – FG [31].

The increase in serum testosterone levels in the second trimester of normal pregnancy in comparison with those values of the 3rd trimester can be attributed to the increase of aromatase activity with progressive course of pregnancy (24) .

A significant increase of the serum TC , TG and VLDL-C Levels in preeclamptic women , can be explained in the following points :

I- The endogenous female sex hormone have significant effect on serum lipid [32]. Oestrogen is responsible for induction of TG

synthesis [33]. There is an increase in the hepatic lipase activity and decrease in lipoprotein lipase activity. Hepatic lipase is responsible for the increased synthesis of the triacylglycerols at the hepatic level, where the decreased activity of lipoprotein lipase is responsible for the decreased catabolism at the adipose tissue level. The net effect of this enzyme will be an increase in circulating triacylglycerol. The second stage of uptake of the remnant of chylomicrons by the liver is delayed so it lead to accumulation of triacylglycerol [32].

2-Serum VLDL increase follows serum TG increase, since the former was calculated from TG values [33]. The increase in triacylglycerol in gestation is estimated mainly in the VLDL, because it is synthesized in the liver and VLDL carries the endogenous triacylglycerol [34].

The same trend of increase in the levels of those constituents were reported in the studies carried out by Demir-SC[35] and suzies – WJ [36] .

In this study , we found a significant increase in LDL-C and decrease in HDL-C in preeclamptic women (2nd and 3rd trimester) These changes can be attributed to : -

1-Increased triacylglycerols play a major role in decreasing HDL-C HDL particles carry cholesterol from peripheral tissues to the largest area of utilization (Liver) and this lead to decrease of HDL-C in serum [37] . There is a direct correlation between adipose tissue lipoprotein lipase activity and plasma HDL-C This direct correlation may be responsible for low levels of HDL- C . Hypertriglyceridemia , leading to low HDL-C mainly due to the actions of cholesteryl ester transfer protein (CETP)(37), which facilitates transfer of cholesteryl ester from HDL to VLDL

, IDL and in exchange for triacylglycerol , relieving product inhibition of LCAT activity in HDL-C. LCAT activity was lower in pregnancy induced hypertension [38].

2-Oestrogens were shown to increase serum HDL- C levels and decrease of LDL-C Levels [39]. Therefore , the low level of HDL-C and a consequent increase in LDL-C level may be attributed to hypoestrogenemia of preeclampsia . It may be also due to insulin resistance in the corresponding patients [40].

3-The decrease in albumin lead to decrease in HDL- C because lysolecithin , one of the products of the lecithin cholesterol acyl transferase (LCAT) reaction , is removed by binding to serum albumin [41].

Our results were in good agreement with the results of Bulter-CL [42] .

The increase in TC, TG , VLDL-C and LDL-C in the third trimester of uncomplicated pregnancy may be attributed to the increased metabolic demand of the fetus with the advancing course of pregnancy [43].

Our results are consistent with the results reported by Cekman-MB [44] and inconsistent with those reported by Tayanta-D [34]who found a significant decrease in the LDL-C level in the third trimester of pregnancy . The inconsistency can be attributed to dietary differences between the studied groups .

In this study, we found a significantly decrease in serum total protein and albumin in women with preeclampsia than in normotensive women with similar gestational age .

This decrease in serum total protein and albumin level in patients and

healthy groups (2nd and 3rd trimester) may be attributed to :-

1-During normal pregnancy the hyperfiltration is largely due to profound resistance reduction in the renal afferent arterioles [45]. In PET both glomerular filtration rate and renal plasma flow decrease by 30% to 40% compared with normal pregnancy of the same duration [46]. The basis for the hypofiltration in PET is largely secondary to structural changes into glomerulous as opposed to constriction of afferent arteriolar system and depression in renal plasma flow , which increasing permeability of glomerulous to protein [47].

2-The protein excretion was approximately four fold higher than that of nonpreeclamptic women [48]. When preeclampsia is accompanied by proteinuria , there is a marked fall in albumin and an increase in α_2 – macroglobulin [49]. It's believed that these changes are a result of urinary loss of the proteins of intermediate molecular weight, with a compensatory unselective increased synthesis of protein in the liver , and retention in the serum of macroglobulins , which are too large to pass through the defective glomerular basement membrane [50]. Metabolic studies have shown that albumin synthesis is significantly greater in preeclampsia than in normal pregnancy , and this is stimulated by the liver due to either decrease in estrogen production or low concentration of albumin in the blood [51].

3-The increase in urinary protein excretion in preeclampsia occurs secondary to alterations in the size and / or charge selectivity of the glomerular filterate [52]. Loss of charge selectivity was likely the primary defect in the glomerular filtration barrier in women with

preeclampsia [47]. Preeclampsia is associated with morphological changes in renal endothelial and mesangial cells have been noted enlarged due to their engorgement with lipid . These lipid – induced changes have recently been named glomerular histopathological endotheliosis [53].

5-Proteinuria lead to hypoalbuminemia , low plasma oncotic pressure and intravascular volume depletion , subsequent under perfusion of the kidney stimulates the renin-angiotensin – aldosterone axis , which causes increased renal sodium and volume retention which to increased extracellular fluid[54]. The extracellular fluid expansion leads to a decrease in serum albumin [55].

Our results were good agreement with these reported by Salako – BL [56].

The decrease in serum total calcium and magnesium in preeclamptic pregnancy compared with control group can be attributed to :

1-During normal Pregnancy , there are many mechanism tend to promote lowering of maternal calcium concentration due to an increase in maternal estrogen production which blocks bone resorption and increases calcium excretion in urine [55]

2-The haemodilution occurs during the last trimester of pregnancy [57]. Jord-R found that were calcium was strong association between serum albumin with systolic and diastolic blood pressure [58]. Because there is a strong correlation between total and ionized serum calcium , one would have to assume that the binding characteristics for calcium and its carrier proteins are abnormal in hypertension [58].

3-The prevalence of magnesium deficiency may be due to the difference in the dietary pattern [57]. The haemodilution could be another factor leading to a higher prevalence of deficiency of magnesium [57].

4-Magnesium exclusively excreted in urine and reabsorbed in proximal convoluted tubules by a process called transport maximum (Tmax) its excretion increase as a filtered load increase above the transport maximum, in women with decrease GFR, the filtered load is more excretion of magnesium in urine [59]. During normal pregnancy, the increase in GFR causing increase in calciuria [55].

5-Magnesium homeostasis is linked with calciuria [60]. Studies from the first elucidated the nature of the effects of calcium and magnesium ions at the neuromuscular junctions [61]. Magnesium competes for prejunctional site with calcium ions , the ions competed with each other , high magnesium concentration inhibit release of acetyl choline (Ach) and high calcium concentration increases of Ach from presynaptic nerve terminal . In sever preeclampsia , there is vasospasm , ischemia as well as cellular hypoxia which may cause reperfusion injury following treatment [61].

6-Magnesium is physiologically antagonist to calcium , it follows that in an attempt to mitigate cellular injury by calcium , there will also be influx of magnesium during reperfusion . This could explain why both calcium and magnesium were reduced in the blood of preeclamptic pregnant women [62].

Our results are in good accordance with the results reported by Sukonpan-K [63] and Sanders- GT [64].

References

- 1-Sheinam-A.Dewhurst,s text book of obstetrics & Gynecology for Postgraduates. 8thed. Graphicraft, Hong Kong. 2007;227.2-Symond-EA and Symond-LM. Essential obstetrics and Gynecology. 4th Churchill livingstonec-philadelphia. 2004;163-168.
- 3-Reynold-C, Mabie -WC and Siba-BM, etal. Preeclampsia:hypertension State of pregnancy.Armeinian maedical network.2005.
- 4-Chan-PD and Hahnson-SM. Current clinical strategies publishing. 6th Laguna Hills. California. 2006.
- 5-Edmond-DK.Dewhurst,s text book of obstetrics & Gynecology for Postgraduates. 6thed. Graphicraft, Hong Kong.1999. 169.
- 6-Ficioglu-C and Kutlu-T. The role of androgens in the aetiology and pathology of preeclampsia. J Obstet Gynecol.2003;23:134-137.
- 7-Buhimschi-CB, Maglorri-L and funai-E,etal. Fractional excretion of Angiogenic factors in women with severe preeclampsia. Am College obstet & Gynecol. 2006;107(5):1103-11130
- 8-John-P. Evalution of prteinuria in pregnant women. Am J obstet & Gynecol.2008;57:611
- 9- Takeuchi-K , Zhang-B and ideishi-M , etal. Influence of age & Hypertension on the association between small artery Compliance & coronary artery disease. Am J Hypertension. 2004;17(12);1188-1191.
- 10- Murray-RK, Granner-DK, Mayes-PA and Rodwell-VW. Harper, illusterated Biochemistry.26thed. Mc Graw-Hill, New York. 2003;440-445.s
- 11- Ojeda-NB, Grigore-O and Yanes-LL, etal. Testosterone contributes to marked elevations in mean arterial pressure in adlt male intrauterine growth restricted offspring. Am J Physiol Regul Integr Comp Physiol. 2007;292:758-763.
- 12-Vasudevan and Sreekuman-S. Text book of biochemistry for medical Students.3rded.Jaypee brothers,New delhi. 2001;417-422,218.13-Guyton-AC and Hall-JE. Text book of medical physiology.11thed.Elsevier Saunders. 2006;855.
- 14-Ding-DC, Hsu-S and Chu-TW. Massive ascites in sever preeclampsia .Taiwanes Jobstet Gynecol. 2005; 44(4): 384-386.
- 15-Kaplan-LA and pesce-AJ. Clinical chemistry,theory, analysis, Correlation. 4thed. Elsever science,New York. 2002;387-348, 760.
- 16-Newman-JC and Amara-S. The pathogenesis of preeclampsia :the Magnesium ischemia hypothesis. Med Hypothesis(England).1993;40(4):250-256.
- 17-Kisters-K,Barenbroka-M and Louwenb-F, et al. Membrane intracellular and magnesium,and calcium concentrations in preeclampsia Am J Hypertension. 2000; 13(7):765-769.
- 18-Hojo-M and August-P.Calcium metabolism in normal & hypertensive Pregnancy.Semin nephrol.1995;15 (6):504-511.
- 19- LopezP. Prevention of preeclampsia with calcium supplementation And itsrelation with L-arginine ,nitric oxide pathway.Braz J Med Biol Res.1996;29(6)731-741.20-Ray-J,Vasuhath-K and Kaura,et al. Calcium metabolism in preeclampsia

International J Gynecol obstet. 1999;66 (3):245-250.

21-Petridou-E, Katasouyanni-K and Spanos-E,etal. Tobacco smoking, Pregnancy,PET estrogens,and birth wieght. Epidemiology. 1999;1:247-250.

22-Iou-SG, Eskandari-M and Dabiri-A. Evaluation of estrogen level of. Women with preeclampsia in third trimester. Medical J Islamic World Academy of Sciences. 2005;15(1):19-22.

23-Reyes-MR, Alvarez-SA and Lazalde-B. Estrogens are potentially the only steroids with an antioxidant role in pregnancy. Acta obstet Gynecol Scand. 2006;85(9):1090-1093.

24-Rebecca-T Postischman-N, Roberts-JM and Ness-R. Maternal serum estrogen and androgen concentrations in preeclamptic and uncomplicated pregnancies. Intern J Epidem. 2003;32: 455-460.

25-Michie-EA. Urinary oestriol excretion in pregnancies complicated by Suspected retarded intrauterine growth, toxemia or essential hypertension.J Obstet Gynecol Br Commonw.

1967;74:896-901.26- Sorensen TK, Williams MA and Zingheim RW.etal. Elevated human chorionic gonadotropin and subsequent pregnancy-induced hypertension. Am J Obstet Gynecol. 1993;169:834-838.

27-Steier-JA, Ulstein-M and Myking-OL. Human chorionic gonadotropin and testosterone in normal and preeclamptic pregnancies in relation to fetal sex. Obstetric gynecol.2002;100:552-556.

28-Barbieri-RL,Makris-A and Randall-RW,etal. Insulin stimulates androgen accumulation in incubations of ovarian

stroma obtained from women with hyperandrogenism. J clin endocrinol. Metab.2005;62:204-210.

29-Bammann-BL, Goulam-CB and Jiang-NS.Androgen production and metabolism in normal pregnancy.Am J Obstet Gynecol. 1980;137(3):293-298.

30-Golmammad-IS, Salari-S and Eskandari-M, etal. Evaluation Of androgen and progesterone levels in women with preeclampsia. Iran J Med Sci. 2005;30(4):186-189.

31-Jasim-FG. Role of sex hormones in preeclampsia. PhD thesis.College of Medicine. Al-Nahrain University. 2008.

32-Patrizia-B, Giancarlo-Tand Francu-E, etal. Lipoprotein metabolism during normal pregnancy. Am J Obstet Gynecol. 1999;181 (2):430-434.

33-Friedewald,WT, Levy-RI and Fredrickson,DS. Estimation of the concentration of LDL-C without the use of preparative ultracentrifuge. Clin chem. 1972;18:499.34-Jayanta-D, Mukhapadhyay-AK,and Saha-PK. Study of serum lipid Profile in pregnancy induced hypertension. Indian J Clin Biochem. 2006;21(12):165-166.

35-Demir-SC, Urunsak-IF and Ozgunen FT,etal. Comparison of lipid Profile in normal and hypertensive pregnant women. Ann Saudi Med. 2004;24(5):382-389.36-Suzie-WJ, Sanchez-SE and Laraburre-G,etal. Plasma lipid concentrations in preeclamptic and normotensive

Peruvian women. Intern J Gynecol Obstet.2008;100(9):1059-1062.37- Aziz-R and Mahboob-T. Preeclampsia and lipid profile. Pak J Med Sci. 2007;23(5):751-754.

- 38-Laukidi-B, Senhadji-L and Merzouk-S, et al. Serum Lecithin:cholesterol acyltransferase activity, HDL2 and HDL3 composition in hypertensive mothers and their small for gestational age Eur J Pediatr. 2008;167(5):525-532.
- 39-Bradley-R and Crook-D. Text book of clinical biochemistry: Metabolic and clinical aspects. 1st ed. Churchill livingstone, London. 1995;413-422.
- 40-Kaja-R, Triikkanen-MJ and Vinnikka-L, et al. Serum lipoproteins, insulin and urinary prostanoid metabolites in normal and hypertensive pregnant women, Obstet Gynecol. 1995;85(3):
- 41- Weinstock-GR, Mayneld-C and Albers-J. Evaluation of quality – Control materials for high density lipoprotein cholesterol Quantitation. Clin Chem. 1991; 27:116-133. 356-359.
- 42-Bulter-CL, Williams-B and Raymond-SM, et al. Maternal plasma lipid concentrations in early pregnancy and risk of preeclampsia Am J Hyper. 2004;17(7):574-581.
- 43-Winny-HT, Khoury-MY, and Takahashi-WH, et al. Assessment of serum lipids in pregnant women aged over 35 years and their relation with preeclampsia. Obstet Gynecol, 2008;6(1):63-67.
- 44-Cekmen-MB, Erbagci-AB and Balat-A, et al. Plasma lipid and lipoprotein concentrations in pregnancy-induced hypertension. Clin Biochemis. 2003;36(7):575-578.
- 45-Jeyabalan-A and Conrad-KD. Renal function during normal pregnancy and preeclampsia. Front Biosci. 2007;1(12): 2425-2437.
- 46-Joaquin-ZM, Armando-OCC and Hugo-MZ. Clinical evolution of hypertension and proteinuria in patients who developed preeclampsia. Inter J Gynecol Obstet. 2005;5(1):1-90.
- 47-Hadunewich-M, Karumanchi-SA and Lafaette-R. Pathophysiology Of the clinical manifestations of preeclampsia. Clin J Am Soc Nephrol. 2007;2:543-549.
- 48-John MD, Homuth-V and Jeyabalan-A, et al. New aspects in the pathophysiology of preeclampsia. J Am Soc Nephrol. 2004; 15:2440-2448.
- 49-Horne CHW, Howie-PW and Goudie-RB. Serum alpha macroglobulin, transferrin, albumin in preeclampsia. J Clin Pathol. 2007;23: 514-516.
- 50-Studd-JW and Bailey-DE. Serum protein changes in the preeclampsia eclampsia syndrome. Inter J Obstet Gynecol. 2008;77(9): 796-801.
- 51-Woods-LL Ingelfinger-JR and Rasch-R. Modest maternal protein restriction fails to program adult hypertension in female rats. Am J Physiol Renal Integr Comp Physiol. 2005;289(4): 1131-1136.
- 52-Brow-MA, Wang-MX and Buddle, et al. Albumin excretory rate in normal and hypertensive pregnancy. Clin Sci. 1994;86(3): 251-255.
- 53-Smean-AI, Bisalla-A and Ekele-BA. Comparison of metabolic syndrome variables among pregnant women with and without preeclampsia. J Nation Med Associ. 2008;100(9): 1059-1062.
- 54-Goldman and Ausiello. Cecil medicine. 23rd ed. Saunders Elsevier. 2007;867.
- 55-Nasser-O and Shurideh. Does serum calcium in preeclampsia and normal pregnancy differ. Saudi Med J. 2001;22(10): 868-871.

56-Salako-BL, Odukagbe-AT and Olayemi-O, etal. Serum albumin, creatinine, uric acid, and hypertension disorder of pregnancy.East African Med J. 2003;80(8):424-428.

57-Pathak-P, Kapoor-SK and Kapil-V, etal. Serum magnesium level among Pregnant women in rural community of haryana state. Eur J Clin Nutrition. 2003;57:1504-1506.

58- Jord-R, Sunsfjard-J and Bonaa-KH. Serum calcium and cardiovascular risk factors and disease .Am heart Assoc.1999;34:484-490.

59-Resnick-LM, Barbagallo-M and Bardicef-M,etal. Cellulr-free magnesium depletion in brain and muscle of normal and preeclampsia pregnancy.Am Heart Associ. 2004;44:322-326.

60-Mehta-R and Petrova. Ionized magnesium and gestational age. Indian J Pediotrics. 2007;74:1025-1028.

61-Idogun-ES, Imarengiaye-CO and Momoh-SM. Extracellular calcium and magnesium in preeclampsia and eclampsia. African J rep Health. 2007;11(2):80-85.

62-Wang-,Cottrel-JE and Kass-IS. Small physiologic changes in calcium and magnesium alter excitability and burst firing of CIA pyramidal cell in rat hippocampal shees. J Neurosurg

anesthesiol. 2004;16(3):201-209.

63-Sukopan-S and Phupong-V. Serum calcium and magnesium in Normal and preeclamptic pregnancy. Obstet Gynecol.2005; 273(1):12-16.

64-Sanders-GT, Huijgen-HJ and Sanders-R. Magnesium in disease: overview with special emphasis on the serum ionized magnesium. Clin Chem Lab Med. 1999;37(11):1011-1033.