INTRODUCTION

Preeclampsia is a pregnancy-specific disorder characterized by hypertension and excess protein excretion in the urine ***(Pennington et al., 2012).*** It occurs after the 20thweek of gestation ***(Sabine et al., 2010).***

Preeclampsia (PE) affects 5% of pregnancies and is a significant contributor to maternal mortality and morbidity ***(Anthony et al., 2011).*** In addition, PE frequently coexists with intrauterine growth restriction (IUGR, also called fetal growth restriction), placental abruption, and the need for iatrogenic preterm delivery, which are additional major causes of adverse outcomes ***(Thilaganathan et al., 2010).***

Despite many years of research, a complete understanding of the molecular pathogenesis of PE is still missing. The placenta plays a key role in the pathogenesis of preeclampsia since the symptoms of PE can occur in molar pregnancies which lack a fetus and the disease disappeared once the placenta is delivered ***(Mikat et al., 2012).***

It has been hypothesized that the placenta releases some unknown factor(s) into the maternal circulation that initiates the disorder of preeclampsia in the mother ***(Nicholas et al., 2010).***

The primary pathogenesis of preeclampsia begins with an abnormality of the placental vasculature resulting in decreased perfusion of the placenta that in turn leads to the release of factors that cause widespread maternal endothelial dysfunction. The endothelial dysfunction leads to multi-organ dysfunction ***(Gleicher 2007; Huang et al., 2008; Young et al., 2010).*** Vascular endothelial dysfunction, which may lead to hypertension due to an imbalance between vasoactive agents and vasodilators, is an important element of this disorder ***(Sattar et al., 1996; Thiele et al., 2004; Üstün et al., 2010).***

Abnormal lipid metabolism could be a key factor in the mechanism leading to endothelial dysfunction in preeclampsia ***(Thiele et al., 2004).***

Human placenta and the fetus may derive energy from mitochondrial fatty acid oxidation.

During pregnancy, energy demand is increased due to both fetal and maternal requirements.Consequently impaired mitochondrial function resulting in abnormal energy production could play a key role in dysfunction of the feto-placental unit ***(Sabine et al., 2010).***

Abnormal placentation, eg, absent or restricted physiological dilatation of the spiral arteries, is a key factor in the pathophysiology of early-onset preeclampsia.In the abnormal placenta of preeclampsia, fatty acid oxidationis also disturbed ***(Thiele et al., 2004).***

Carnitine is essential for the transport of fatty acids over the inner and outer mitochondrial membranes into the mitochondrion, where B-oxidation takes place ***(Thiele et al., 2004).***

Reduced capacities of fatty acid oxidation for example lead to the accumulation of acyl-CoA-esters and acylcarnitines ***(Sabine et al., 2010).*** Acylcarnitines are known to have unspecific cytotoxic effects ***(Ter Veld et al., 2009).***

Inhibition of oxidative phosphorylation by (long chain) acyl intermediates resulting from impaired fatty acid oxidation might result in the production of reactive oxygen species (ROS) and enhanced lipid peroxide formation, which could lead to endothelial damage ***(Sabine et al., 2010).***

Mitochondrial damage could result as a consequence of lipid peroxides and their related free radicals produced by the PE/HELLP feto-placental unit.Resulting toxic metabolic precursors are likely to be transferred to the maternal circulation causing tissue damage associated with PE ***(Sabine et al., 2010).***

AIM OF THE WORK

To evaluate the possible role of abnormal fatty acid oxidation as expressed by elevated plasma carnitine levels in preeclampsia pathogenesis.

PREECLAMPSIA

Preeclampsia is a pregnancy-specific disorder characterized by hypertension and excess protein excretion in the urine ***(Pennington et al., 2012).*** It occurs after the 20thweek of gestation ***(Sabine et al., 2010).***

Preeclampsia is defined according to ACOG as:

Hypertension: diastolic blood pressure> 90 mmHg, systolic blood pressure >140 mmHg based on at least 2 measurements taken at least 4 hours apart

Significant proteinuria: Proteinuria >300 mg\24 hours or at least +2 on dipstick ***(ACOG, 2010).*** Absence of either hypertension or proteinuria excludes the diagnosis ***(NICE, 2010).***

In the past, edema was considered a diagnostic criterion but recently it has been eliminated as a requirement for diagnosis ***(Hals and Crump, 2000).***

Preeclampsia (PE) affects 5% of pregnancies and is a significant contributor to maternal mortality and morbidity. The effects of PE extend beyond pregnancy, with associated risk of type II diabetes, hypertensive disorders, and coronary artery disease in later life ***(Bellamy et al., 2007; Anthony et al., 2011).***

Preeclampsia is a common complication of pregnancy with potentially devastating consequences to both the mother and the baby. It is the leading cause of maternal deaths in developing countries. In developed countries it is the major cause of iatrogenic premature delivery and contributes significantly to increasing health care cost associated with prematurity ***(Silasi et al., 2010).***

In addition, PE frequently coexists with intrauterine growth restriction (IUGR, also called fetal growth restriction), placental abruption, and the need for iatrogenic preterm delivery, which are additional major causes of adverse outcomes ***(Thilaganathan et al., 2010).***

There is currently no known treatment for preeclampsia; ultimate treatment involves delivery of the placenta ***(Silasi et al., 2010).***

Hypertensive disorders during pregnancy:

Due to gestational physiology, blood pressure (BP) may decrease during the first trimester of pregnancy, reaching its nadir by mid-pregnancy. A normal BP of 110-120/70-80 mmHg in a healthy woman of 20-30 years of age might decrease by 5-10 mmHg during this interval, as compensatory increases in blood volume and vasodilatation occur. systolic blood pressure (SBP) is less affected than diastolic blood pressure(DBP) because of the increased cardiac output that offsets the systemic vasodilation. Blood pressure then usually returns to preconception levels during the third trimester ***(Marvin et al., 2012).***

Hypertensive disorders during pregnancy occur in women with pre-existing primary or secondary chronic hypertension, and in women who develop new-onset hypertension in the second half of pregnancy ***(Kang and Struben, 2008).***

Chronic hypertension:

Is hypertension that is present at the booking visit or before 20 weeks or if the woman is already taking antihypertensive medication when referred to maternity services. It can be primary or secondary in etiology.

Gestational hypertension:

Is new hypertension developing after 20 weeks without significant proteinuria.

Preeclampsia:

Is new hypertension developing after 20 weeks with significant proteinuria.

Severe preeclampsia:

Is preeclampsia with severe hypertension (systolic blood pressure >160 mmHg, diastolic blood pressure >110 mmHg based on at least 2 measurements taken at least 4 hours apart) and/or with symptoms, and/or biochemical and/or haematological impairment ***(NICE, 2010).***

Eclampsia:

Is the most dangerous complication of preeclampsia, characterized by generalized tonic-clonic convulsion, and may develop before or after delivery. This form of preeclampsia is associated with higher maternal and fetal mortality.

HELLP syndrome [hemolysis (H), elevated liver enzymes (EL), and low platelets (LP)]:

Is a severe progressive course of preeclampsia ***(Kang and Struben, 2008).***

Classification of preeclampsia:

Although more than 100 years have passed since preeclampsia was first described, knowledge of its etiology and pathophysiology is limited. By regarding preeclampsia as one disease entity, the issue may be confused. The heterogenous pre-eclamptic syndrome should be considered a result of

different pathogenic processes, and some investigators have suggested a distinction between maternal and placental etiologic factors. Furthermore, by stratifying cases of preeclampsia according to clinical subtypes, important associations may be revealed that otherwise could go unnoticed. Thus, the differential pattern that was observed between various risk factors on the one hand, and clinical subtypes on the other, may support the suggestion that a useful approach to clinical studies of preeclampsia is to divide the syndrome into appropriate subgroups. Based on recent observations, relevant subgroups for future studies could be established based on genetic characteristics (maternal, paternal or both) or placental histopathology ***(Rønnaug et al., 2005)*.**

Classification of severity:

American Congress of Obstetricians and Gynecologists (ACOG): classification of severity

Disease severity is based on the BP measurement and whether there are signs of systemic involvement.

- Mild-moderate
  - BP is 140 to 159 mmHg systolic and/or 90 to 109 mmHg diastolic.
- Severe
  - If one or more of the following criteria are present:
    - BP is ≥160 mmHg systolic and/or ≥110 mmHg diastolic (on 2 occasions at least 6 hours apart, while the patient is on bed rest)
    - Proteinuria of ≥5 g/24 hours or ≥3+ (on 2 random urine samples, collected at least 4 hours apart)
    - Oliguria <500 mL/24 hours
    - Cerebral or visual disturbances

- Pulmonary edema or cyanosis
- Epigastric or right upper quadrant pain
- Impaired liver function
- Thrombocytopenia
- Fetal growth restriction

HELLP syndrome is a subtype of severe preeclampsia characterized by hemolysis (H), elevated liver enzymes (EL), and low platelets (LP) ***(ACOG, 2002).***

National Institute for Health and Care Excellence 2010 (NICE, UK): classification of severity:

Severity of disease is based on BP measurement alone:

- Mild: BP is 140 to 149 mmHg systolic and/or 90 to 99 mmHg diastolic
- Moderate: BP is 150 to 159 mmHg systolic and/or 100 to 109 mmHg diastolic
- Severe: BP is ≥160 mmHg systolic and/or ≥110 mmHg diastolic.

(NICE, 2010)

Classification of onset:

Preeclampsia has been characterized by some investigators into 2 different disease entities:

a) Early-onset preeclampsia: is usually defined as preeclampsia that develops before 34 weeks of gestation.

b) Late-onset preeclampsia: develops at or after 34 weeks of gestation.

(Raymond et al., 2011)

Pathophysiology of Preeclampsia:

The pathophysiology of preeclampsia likely involves both maternal and fetal/placental factors. However, the molecular mechanisms behind preeclampsia are not clear ***(Young, 2013).***

It has been reported that the presence of genetically determined inborn errors of long-chain fatty acid oxidation (FAO) in the fetus predisposes the mother to suffer from PE. Incidences of PE are much higher compared to those of inborn errors of metabolism affect in fatty acid oxidation (FAO). Based on this discrepancy, it was postulated that compromised mitochondrial function caused by other factors than inborn errors of fatty acid oxidation (FAO) may be involved in this obstetric disease ***(Sabine et al., 2010).***

The possible role of abnormal lipid metabolism, inparticular, abnormal fatty acid oxidation, in the pathophysiologyof preeclampsia was recently demonstrated:

79% of the pregnant women who are carrying a fetus with a long-chain 3-hydroxyacyl-CoA dehydrogenase deficiency, a defect in the mitochondrial fatty acid oxidation, develop the hemolysis, elevated liver enzymes, low platelets (HELLP) syndrome, a severe consequence of preeclampsia ***(Thiele et al., 2004).***

This is most likely the result of toxic metabolites originating from the long-chain 3-hydroxyacyl-CoA dehydrogenase– deficient placenta entering the maternal circulation. This finding is also of interest because it is generally accepted that the placenta is a key factor in the pathophysiology of preeclampsia and is thought to be the main source of factors that eventually lead to endothelial cell dysfunction in the mother ***(Robertset al., 2002; Redman et al., 2003; Thiele et al., 2004).***

The presence of elevated lipid hydroperoxides, reduced antioxidants, and dyslipidemia suggests that abnormalities in the fatty acid metabolism exist in preeclampsia ***(Thiele et al., 2004).***

Also early supplementation with antioxidants (vitamins C and E) may be effective in decreasing oxidative stress and preventing preeclampsia as shown in a subgroup of women identified by uterine artery Doppler screening ***(Thiele et al., 2004).***

To explain the pathogenesis of preeclampsia, there are several hypotheses including altered angiogenic balance, circulating angiogenic factors like marinobufagenin (a bufadienolide trigger), and activation of the renin-angiotensin system ***(Young, 2013).***

Epigenetically-modified circulating cell-free nucleic acids in plasma and serum could be novel markers with promising non-invasive clinical applications in the diagnosis of preeclampsia ***(Young, 2013).***

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Abnormal development of the placenta:

Because the placenta is central to preeclampsia pathogenesis, researches have focused on the association between abnormal placental vascular development and the development of preeclampsia ***(Young, 2013).***

In normal pregnancies, extravillous cytotrophoblasts of fetal origin invade the uterine spiral arteries of the decidua and myometrium. These invasive cytotrophoblasts replace the endothelial layer of the maternal spiral arteries, transforming them from small, high-resistance vessels into large-caliber vessels ***(Young, 2013).***

In preeclampsia, this transformation is incomplete. Cytotrophoblast invasion of the spiral arteries is limited to the superficial decidua and does not reach the myometrium **(Figure 1)** ***(Young, 2013).***

Defective differentiation of trophoblasts might be an explanatory mechanism for defective trophoblast invasion of the spiral arteries in preeclampsia ***(Young, 2013).***

For trophoblast differentiation to occur there must be alterations in the expression of a number of different classes of molecules, including cytokines, adhesion molecules, extracellular matrix molecules, metallo-proteinases, and the class IB major histocompatibility complex molecule, histocompatibilty leukocyte antigen-G ***(Young, 2013).***

During normal pregnancies, invading trophoblasts alter their adhesion molecule expression from one set of endothelial transmembrane receptors (integrin alpha6beta1, integrin alphavbeta5, and E-cadherin) to another (integrin alpha1beta1, integrin alphavbeta3, and VE-cadherin) ***(Young, 2013).***

Hypoperfusion, hypoxia, and ischemia are critical components in the pathogenesis of preeclampsia because the hypoperfused placenta elaborates many factors into maternal vessels that alter maternal endothelial cell function and lead to the systemic symptoms of preeclampsia ***(Young, 2013).***

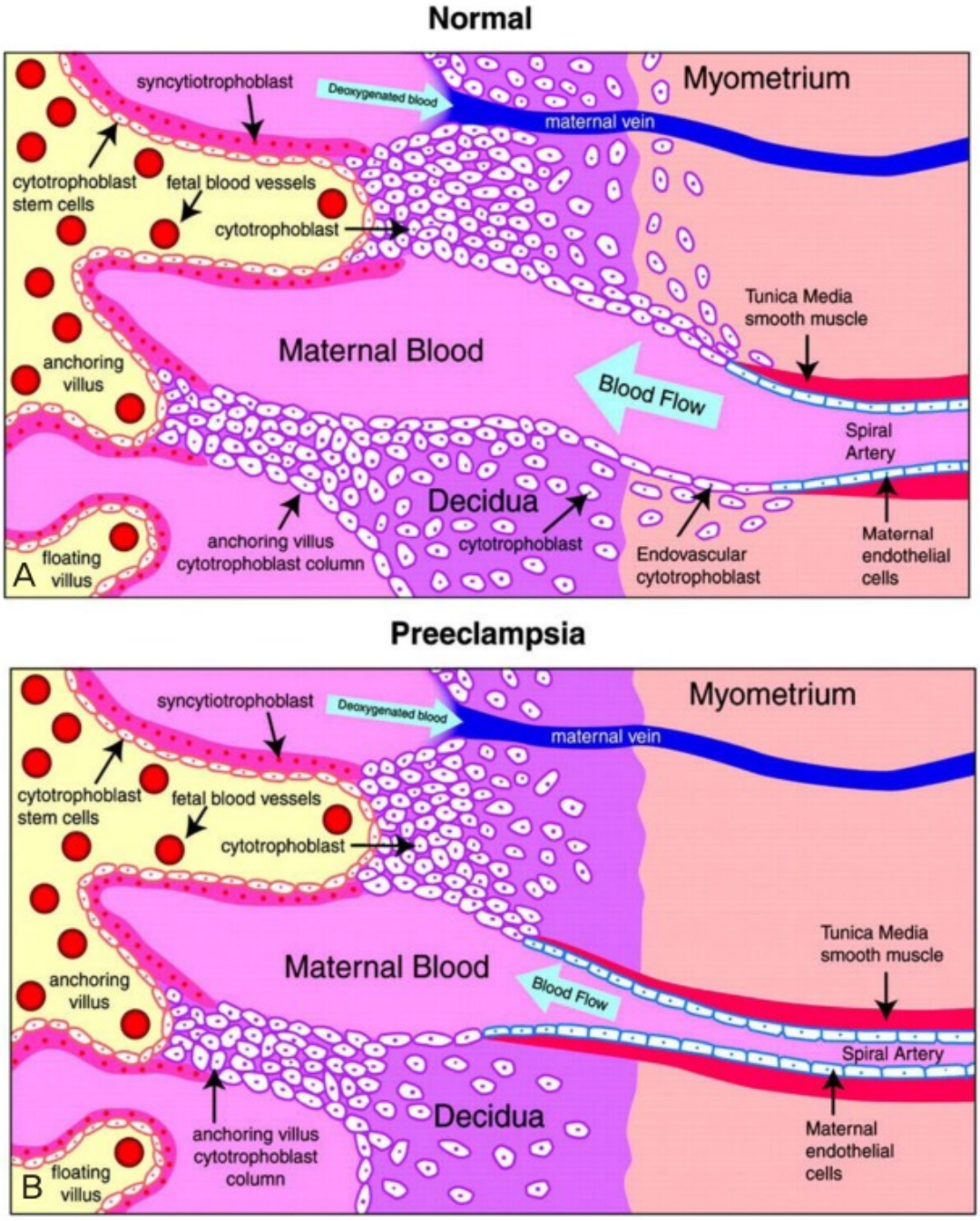

Figure (1): Abnormal placentation in preeclampsia. In normal pregnancies, extravillous cytotrophoblasts of fetal origin invade the uterine spiral arteries of the decidua and myometrium. These invasive cytotrophoblasts replace the endothelial layer of the maternal spiral arteries, transforming them from small, high-resistance vessels into large-caliber vessels **(A).** However, in preeclampsia, this transformation is incomplete. Cytotrophoblast invasion of the spiral arteries is limited to the superficial decidua and does not reach the myometrium **(B)** ***(Lam et al., 2005).***

Altered Angiogenic Balance and Circulating Angiogenic Factors:

Many angiogenic factors are produced by the human placenta. The most important factors are vascular endothelial growth factor (VEGF) and placental growth factor (PlGF) ***(Young, 2013).***

VEGF is an endothelial-specific mitogen that plays a key role in promoting angiogenesis .VEGF's activities are mediated by its interaction with two high-affinity receptors-tyrosine kinase-kinase-insert domain region (the kinase domain region or vascular endothelial growth factor receptor-2) and fms-like tyrosine kinase 1 (flt-1) ***(Vitoratos et al., 2012).***

PlGF is an angiogenic growth factor that is thought to amplify VEGF signaling by displacing VEGF from the flt-1 receptor and allowing it to bind to the more active kinase-insert domain (KDT) receptor. Increased sFlt-1 during preeclampsia is associated with decreased free VEGF and free PlGF in the blood ***(Vitoratos et al., 2012).***

Soluble endoglin is an antiangiogenic protein. Circulating soluble endoglin levels increased markedly beginning from two to three months before the onset of preeclampsia ***(Vitoratos et al., 2012).***

An increased level of soluble endoglin was associated with an increased endoglin/sflt-1 ratio ***(Levine et al., 2006).***

There is substantial evidence that effective angiogenesis requires the synthesis of bioactive endothelium-derived nitric oxide (NO) .A number of angiogenic factors up-regulate the endothelial expression of NO synthase (NOS) stimulating the release of endothelium-derived NO. NOS is partially regulated by negative feedback from NO, but there are other important inhibitors that are endogenously present in humans, such, the competitive inhibitor asymmetric dimethylarginine (ADMA).There are some conflicting findings concerning ADMA concentrations in pregnancies complicated by preeclampsia. However, ***Kim et al. (2011)***

suggested that increased maternal circulating ADMA levels, a higher expression of placental eNOS protein, and a lower expression of placental phospho-eNOS protein contribute to the development of preeclampsia.

Marinobufagenin, a Bufadienolide Trigger of Preeclampsia Pathogenesis:

Presently, the key biomarkers of the syndrome associated with preeclampsia pathogenesis are thought to be marinobufagenin (MBG) and angiogenic imbalance ***(Uddin et al., 2012).***

Data from a rat model that mimics human preeclampsia showed that urinary excretion of MBG increased before the onset of hypertension and proteinuria, and that affected animals have increased vascular leakage and blood-brain barrier permeability ***(Uddin et al., 2012).***

The cardenolides and bufadienolides are group of steroid compounds, which belong to a class of circulating substances called "cardiac glycosides" **(Figure 2)**. Cardiac glycosides are natriuretic and cause vasoconstriction, which can lead to hypertension ***(Young, 2013).***

The bufadienolides are more important in diseases such as preeclampsia than the cardenolides. The best studied of the bufadienolide compounds is MBG **(Figure 2A)**, an endogenous vasoconstrictor mammalian cardiotonic bufadienolide ***(Young, 2013).***

Preeclampsia is thought to result from inadequate placentation, related to a failure of the trophoblasts to adequately remodel the vasculature of the uterus. This leads to hypoperfusion of the placenta, often causing oxidative stress, and endothelial dysfunction, which are related to the development of the preeclampsia syndrome. MBG interferes with proliferation, migration, and the invasive capacity of the cytotrophoblasts and has deleterious effects on human cytotrophoblast cell function, which suggests a role for MBG in the abnormal placentation and altered vascular function of preeclampsia ***(Young, 2013).***

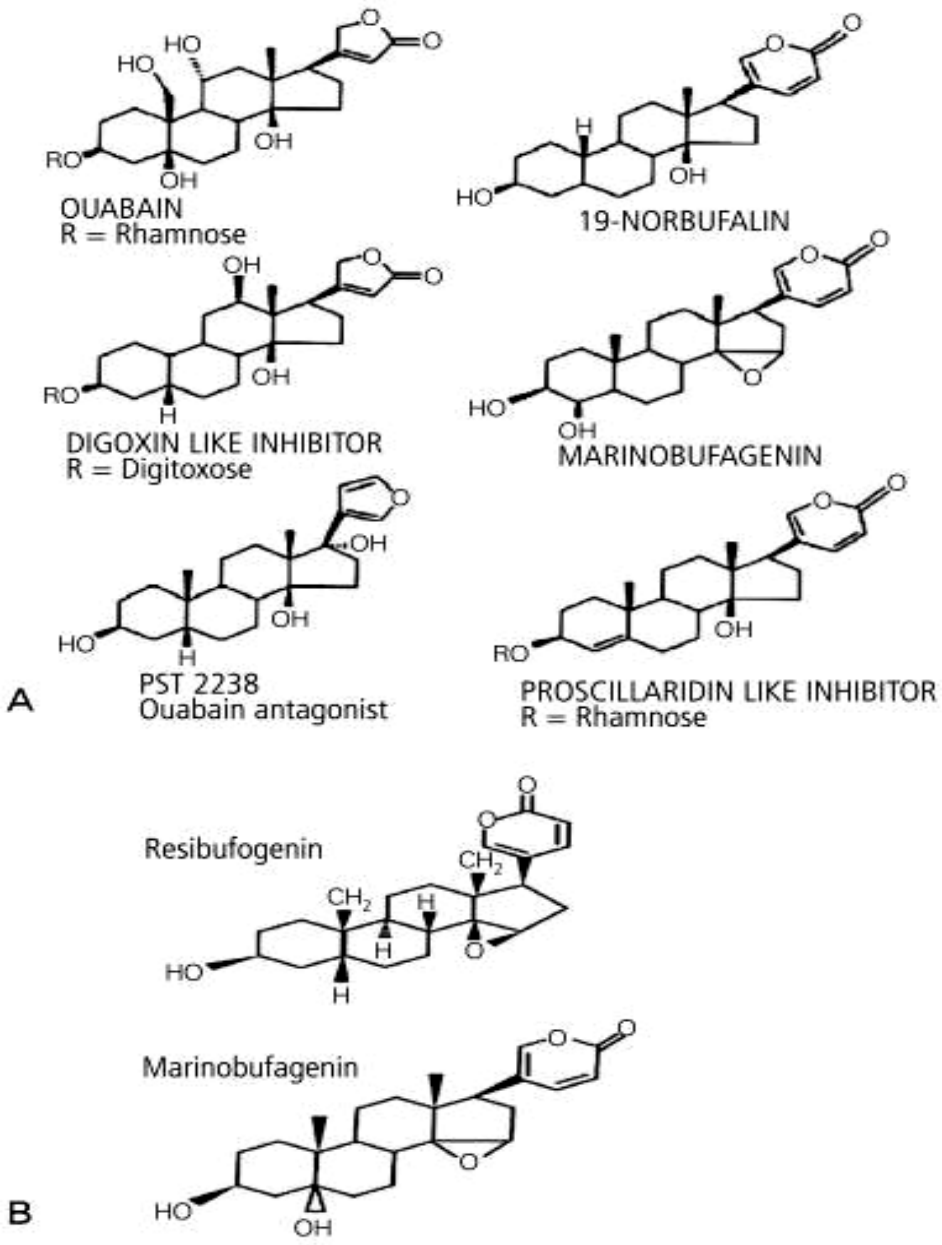

Figure (2): (A) Chemical structures of the bufadienolides and the cardenolides. The compounds on the left side are cardenolides and those on the right side are bufadienolides. **(B)** The chemical structures of marinobufagenin and resibufogenin ***(Uddin et al., 2011).***