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PREECLAMPSIA AND STERILE INFLAMMATORY MOLECULES

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Abstract
Preeclampsia (PE), as a pregnancy-specific diseases, has become one of the main causes of maternal and fetal morbidity & mortality worldwide. It is also one of the major risk factor for pre term birth. PE is typically characterized by hypertension, proteinuria, and an excessive maternal systemic inflammatory response. Recent evidences suggests the notion that natural killer T (NKT) cells (a small, but significant immunoregulatory T cell subset of human peripheral blood lymphocytes) play pivotal roles in pregnancy. NKT cells with unique transcriptional and cytokine profiles exist in different peripheral tissues acting as mediators between the innate and adaptive immune systems. NKT cells secrete Interleukin-4 (IL-4) and Interferon-γ (IFN-γ) which might regulate the balance between Type 1 T helper (Th1) and Type 2 T helper (Th2) responses. Sterile inflammatory molecule and damage-associated molecular pattern (DAMP) released from various cells during stress has been implicated in inflammation. Studies showed that there is a direct relationship between inflammation and preeclampsia. Here, we intend to summarize the concept of the emerging link between sterile inflammatory molecules and PE. Further more, we will discuss the possible therapeutic strategies that target sterile inflammatory molecules for the PE.

Keywords
PE, sterile inflammatory molecules, circulating nucleicacids, Decorin.

Introduction

Preeclampsia (PE) is a pregnancy-specific hypertensive disorder characterized by hypertension, proteinuria and other systemic disturbances at or after 20 weeks of gestation (1). It is one of the leading causes of maternal and fetal major adverse events including death and iatrogenic pre term birth (2). The precise mechanisms of PE's pathogenesis, early and accurate diagnosis of this disease remains a challenge till date. It is widely acknowledged that the placenta is the central organ in the pathogenesis, and PE is caused by maternal responses to Placentation and associated with an increased inflammatory state. It has been further proposed that PE is an excessive maternal inflammatory response to pregnancy, involving pro-inflammatory cytokines such as tumor necrosis factor, inter leukin and HMGB1 (3). A recent study reported a presence of maternal adverse events including eclampsia without any traditional clinical and laboratory findings such as hypertension and proteinuria. Hence, there is an urgent need to investigate practical, routinely used, and inexpensive markers for early and accurate diagnosis and risk stratification of PE.

PE presents as gestational hypertension (systolic ≥ 140mmHg or diastolic ≥ 90 mmHg) and signs such as proteinuria, renal insufficiency, thrombocytopenia, hepatic dysfunction, and pulmonary edema (4). This can result in increased morbidity and mortality rates in affected mothers

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and fetuses, if left un treated. The only known treatment for PE is the delivery of both fetus and the placenta, often resulting in premature birth, leading to deleterious consequences. Since the treatment for PE involves removing the placenta, many studies have thought that an abnormal placenta plays a causal role in the pathogenesis of PE (5-7).

PE affects 2-8% of pregnancies (8) with eclampsia accounting for 50,000 casualties per year worldwide. It is known that a fetus does not have to be present for pre-eclampsia to develop, as this disorder arises in cases of molar pregnancies within creased placental size and no fetus(9). PE is a major cardiovascular risk for the duration of the pregnancy, post-parturition and in later life, while also being related to maternal malfunction of the vasculature (10-11). Also, endothelial dysfunction may contribute to elevate the peripheral resistance of blood vessels, which forms an essential component of the maternal syndrome (12-13). Although the etiology of pre-eclampsia is still poorly understood, a deficient supply of blood to the placenta giving rise to abnormal placentation is considered to be one of the main features of the disease. Inadequate placentation and poor perfusion are thought to provoke the secretion of many anti-and pro-angiogenic mediators, as well as inflammatory cytokines in to the maternal vasculature.

Inflammation is essential for successful female reproduction. Inflammatory processes are implicated in every step of fertility from the menstrualcycle (ovulation, menses) to early pregnancy (implantation, decidualization) and later during labor (myometrial activation, cervical ripening, weakening of fetal membrane), whereas quiescence of these mechanisms is maintained by local immune cells during gestation to allow maternal tolerance of fetal antigens allograph. However, untimely inflammatory triggers shifting the immunological balance towards activation can lead to adverse pregnancy out comes including preterm birth.

Failure to mount a local inflammatory response in early or late gestation can also lead to adverse conditions, including miscarriages. Evidence shows that impaired inflammatory response is implicated in numerous female reproductive tract pathologies including menstrual disorders(14), endometriosis-associated infertility (15), recurrent miscarriage (16-17), intrauterine growth restriction (18),PE (19-20) and preterm labor (21-22). Infertility has an estimated global prevalence of 9% with >72 million infertile women worldwide (23), whereas preterm birth and PE, the two leading causes of perinatal mortality and morbidity, have an estimated prevalence of>11% (24) and 3-5% (25-26), respectively. Therefore, understanding the mechanisms where inflammation is untimely triggered in the uterus is primordial to develop effective therapeutics to improve fertility and decrease poor obstetrical outcomes.

Inflammation linked reproductive disorders do not require infections. A majority of patients suffering from PE, preterm labor or other inflammatory diseases during pregnancy display no clinical signs of infection either. Although observational and causal data accumulated for over30 years have linked infection to preterm labor, preterm birth without infection is more prevalent (27). Furthermore, antibiotics are inefficient to prevent preterm labor in women with infection (28),suggestingthatinfection-induced pro labor effects arise from inflammation (self) rather than infection (non-self).

The development of PE is typically a *"two-stage"* process (29): the first stage is the incomplete remodeling of spiral arteries leading to poor placentation and the second stage is the resulting stress, especially hypoxia-induced oxidative stress (30-31), leading to the symptoms typical of late-stage PE. However, the actual originating causes of either of these two stages essentially remains obscure, despite the emergence of many theories. Thus, PE is also known as the*"disease of theories"*(32).

Normal pregnancy is associated with intensive changes in the maternal cardiovascular system that enables adequate oxygen delivery and nutritive ingredients to the fetus. Physiological vascular adaptation (increased blood volume, increased cardiac minute volume and reduced vascular resistance) is followed by increased endogenous production of nitric oxide (NO) and improved response of smooth muscles on the reaction of NO (33-34). Impaired response of the blood vessels on vasoconstrictor agonists during the pregnancy can be partly regulated by NO (35). Recently, several *in vitro* studies assessing the role of NO in vascular reactivity in pregnancy and PE, observed that the biosynthesis of NO increased during pregnancy, especially in the second trimester, with its peak in the third trimester (36-38). However, some studies showed that there were no changes in the biosynthesis of NO in normal pregnancy compared with non-pregnantwomen (39-40) suggesting that the biosynthesis of NO during normal pregnancy still remains controversial.

Risk Factor of PE

PE occurs with increased frequency among young, nulliparous women. However, the frequency distribution is bimodal, with a second peak occurring in multiparous women greater than 35 years of age. A number of maternal risk factors have been recognized to identify high-risk pregnant women, including preconception obesity, chronic hypertension, family history, and more. It is hypothesized that an increase in adipose tissue, which is a rich source of pro-inflammatory cytokines and complement proteins, causes an aggravated systemic inflammatory response, angiogenic imbalances in circulation and the placenta, and abnormal placental development resulting in PE (41). Lykke et al. found that PE, spontaneous preterm delivery, or fetal growth deviation in a first single ton pregnancy predisposes women to those complications in their second pregnancy, especially if the complications were severe (42).

In a cohort study of 536,419 Danish women, delivery between 32 and 36 weeks of gestation increased the risk of PE from 1.1% to 1.8% in the second pregnancy, whereas delivery before 28 weeks increased the risk of PE to 3.2%. PE in a first pregnancy, with delivery between 32 and 36 weeks of gestation, increased the risk of PE in a second pregnancy from 14.1%to 25.3% (42). Some risk factors contribute to poor placentation, where as others contribute to increased placental mass and poor placental perfusion secondary to vascular abnormalities. In addition to those discussed above, PE risk factors also include some disorders such as hydatidi form mole, agestation altrophoblastic disease, thrombophilia, urinary tract infection, diabetes mellitus, collagen vascular disease, etc.

Some studies have referred to obesity as arisk factor for PE and showed that the relationship between maternal weight and PE is a progressive risk and varies from 4.3% in women with a BMI<19.8, up to13.3% for women with a BMI ≥ 35 (43).

Signs & Symptoms of PE

PE can develop without any presenting symptoms or show up as swelling and/or edema, proteinuria and high blood pressure. Other symptoms include, headache, nausea and vomiting, abdominal and shoulder pain, lower back pain, blurring of vision, hyperreflexia, and shortness of breath with anxiety.

PE is a serious condition of pregnancy, and can be particularly dangerous because many of the signs are silent while some symptoms resemble the normal effects of pregnancy. The disease is sometimes referred to as a silent killer because most women do not realize that their blood pressure is going up until it's too late.

High blood pressure (Hypertension)

High blood pressure during pregnancy is one of the indicators that PE may be developing. It is traditionally defined as blood pressure 140/90 or greater, measured on two separate occasions atleast four hours apart. During pregnancy, arise in the diastolic blood pressure by 15 degrees or more, or a rise in the systolic blood pressure by 30 degrees or more can be a cause for concern.

Proteinuria

In PE, when the urine contains proteins, due to the loss of kidney function, it is termed as proteinuria.

Nausea or Vomiting

These are significant when the onset is sudden and after mid-pregnancy. Morning sickness usually disappears after the first trimester, so the sudden appearance of nausea and vomiting after mid pregnancy may be linked to PE.

Changes in Vision

Vision changes are one of the most serious symptoms of PE. They may be associated with central nervous system irritation or be an indication of swelling of the brain(cerebral edema). Common vision changes include sensations of flashing lights, auras, light sensitivity, or blurry vision or spots.

Stages of PE

For many years PE has been considered to be at wo-stage disease. The first stage comprises poor placentation and the second stage is the clinical expression of the disease i.e., hypertension and proteinuria. Stage 1 is preclinical and without symptoms, occurring between 8-18 weeks of pregnancy when the uteroplacental circulation is established by spiral artery remodeling. This results in dysfunctional perfusion of placental intervillous space with oxidative and hemodynamic stress molecules. The damaged placenta releases excessive pro-inflammatory and anti-angiogenic factors into the maternal circulation.

The precursors of poor placentation are immunological reactions generated from the adaptability of the mother's immune system to the foreign entity i.e., the fetus. Placentation begins after 8 weeks when the uteroplacental circulation, previously closed by tropho blast plugs in the spiral arteries, begins to open. Defective placentation may arise from premature opening, and perfusion of the intervillous space by oxygenated arterial blood. When defective placentation extends over about 10 weeks, it presents as PE. The second half of pregnancy is associated with excessive or deficient placental derived factors in the mother's blood, secondary to placental damage, before the appearance of clinical signs. Superimposition of a second and later spiral artery lesion, called acute a therosis further reduces utero placental perfusion and predisposes spiral artery thrombosis, which underlies the occurrence of placental infarcts and hence PE (44).

Mild PE

Women with mild PE are hospitalized for further evaluation and, if indicated, a pretermdelivery would be necessary. A

CLASSIFICATION OF PE

Mild Preeclampsia	Severe Preeclampsia
Blood pressure ≥140 mm Hg systolic or ≥90 mm Hg diastolic but <160/110 mm Hg	Blood pressure ≥160 mm Hg systolic or ≥110 mm Hg diastolic on 2 occasions at least 6 hours apart while the patient is on bed rest
Proteinuria ≥300 mg/24 h but <5 g/24 h	Proteinuria of 5 g or higher in 24-hour urine specimen or 3+ or greater on 2 random urine samples collected at least 4 hours apart
Asymptomatic	Oliguria <500 mL in 24 hours Cerebral or visual disturbances Pulmonary edema or cyanosis Epigastric or right upper quadrant pain Impaired liver function Thrombocytopenia Fetal growth restriction

Figure 1: (Current Diagnosis & Treatment Obstetrics & Gynecology, 11th Ed. 26. Hypertension in Pregnancy by David A. Millwe, MD)

complete blood count and levels of serum transaminases, lactate dehydrogenase, and uric acid should be checked weekly to twice weekly. Delivery is indicated if the cervical status becomes favorable, antepartum testing is abnormal, the gestational age reaches 40 weeks, or evidence of worsening PE is seen. Women with mild PE before 37 weeks' gestation are evaluated with bed rest, twice-weekly antepartum testing, and maternal evaluation as described (45). Corticosteroids are administered if the gestational age is <34 weeks; amniocentesis is performed as needed to assess fetal pulmonary maturity. Any evidence of disease progression constitutes an indication for hospitalization and consideration of delivery. The benefit of prophylactic in trapartum magnesium sulfate in preventing convulsions in patients with mild PE has not been conclusively demonstrated (46).

Severe PE

Severe PE mandates hospitalization. Delivery is indicated if the gestational age is 34weeks or greater, fetal pulmonary maturity is confirmed, or evidence of deteriorating maternal orfetal status is seen. Acute blood pressure control may be achieved with hydralazine, labetalol, ornifedipine. The goal of antihypertensive therapy is to achieve a systolic blood pressure <160 mmHg and a diastolic blood pressure <105 mm Hg (47). Four large randomized controlled trials comparing magnesium sulfate with other methods of treatment to prevent convulsions in women with severe PE have demonstrated that magnesium sulfate is associated with a significantly lower rate of eclampsia than either no treat mentor nimodipine. Lucas and colleagues reported no seizures among 1049 preeclamptic women receiving magnesium sulfate prophylaxis. Expectant management is contraindicated in the presence of fetal compromise, uncontrollable hypertension, eclampsia, DIC, HELL Psyndrome, cerebraledema, pulmonaryedema, or evidence of cerebral or hepatic hemorrhage. When severe PE is diagnosed before 24 weeks of gestation, the likelihood of a favorable outcome is low (48).

Pathogenesis of PE

The pathogenesis of PE is not fully elucidated but much progress has been made in the last decades. The placenta has always been a central figure in the etiology of PE because the removal of the placenta is necessary for symptoms to regress. Pathologic examination of placenta from pregnancies with advanced PE often reveals numerous placental infarcts and sclerotic narrowing of arterioles. The hypothesis that defective trophoblastic invasion with associated uteroplacental hypo perfusion may lead to PE is supported by animal and human studies (49). Thus, a two-stage model was developed: incomplete spiral artery remodeling in the uterus that contributes to placental ischemia (stage 1) and the release of antiangiogenic factors from the ischemic placentain to the maternal circulation contributes to endothelial damage (stage2) (Figure 2).

During implantation, placental trophoblasts invade the uterus and induce the spiral arteries to remodel, while obliterating the tunica media of the myometrial spiral arteries; this allows the arteries to accommodate increased blood flow independent of maternal vasomotor changes to nourish the developing fetus (50). Part of this remodeling requires that the tropho blasts adopt anendothelial phenotype and its various adhesion molecules. If this remodeling is impaired, the placenta is likely to be deprived of oxygen, which leads to a state of relative ischemia and an increase inoxidative stress during states of intermittent perfusion. This abnormal spiral arteryre modeling has been shown to be the central pathogenic factor in pregnancies complicated by intrauterine growth restriction, gestational hypertension, and PE. However, one limitation to this theory is that these findings are not specific to PE (51).

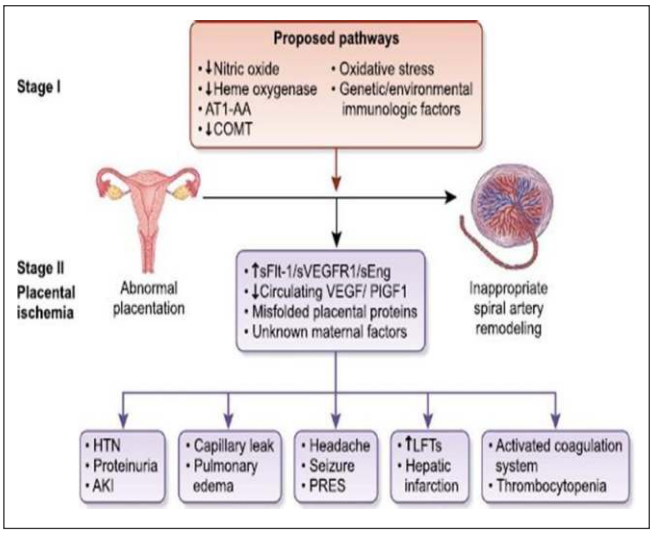

Figure2: 2-stage model of PE pathogenesis.

Angiogenic Factors

In 2003, Maynard et al. showed that soluble fms–like tyrosine kinase 1 (sFlt-1) was up-regulated in the circulation of preeclamptic women. sFlt-1 is a splice variant of the vascular endothelial growth factor (VEGF) receptor, which does not contain the cytoplasmic and membrane domains of the receptor. This allows it to circulate and bind to VEGF and placental growth factor (PlGF), essentially antagonizing their binding to cell surface receptors. When sFlt-1 was injected into rats using an adeno virus, they developed significant hypertension and albuminuria and histologic changes consistent with PE (i.e., glomerular enlargement, endotheliosis, and fibrinde position within the glomeruli). Thus, sFlt-1 seems to be a key mediator in the development of PE (52).

A second placenta–derived protein, soluble endoglin (sEng), was also found to be upregulated in PE. sEng, a circulating coreceptor of TGF-β, can bind to TGF-β in the plasma. Antagonizing TGF-β, a proangiogenic factor, is analogous to

sFlt-1 antagonizing VEGF. In fact, elevated levels of sEngin the circulation have been shown to induce signs of severe PE in pregnant rats (53). The true significance of these angiogenic markers and their ability to predict adverse maternal or fetal outcomes, showed that in a group of women with the clinical diagnosis of PE an elevated level of sFlt-PlGF ratio (angiogenic form) is associated with worse maternal and fetal out comes compared with in women with a lower ratio (non angiogenic form).

Nitric Oxide Pathway

The nitric oxide (NO)/nitric oxide synthase (NOS) system is also disrupted in PE. NO is a potent vasodilator that acts to induce relaxation in vascular smooth muscle cells via a c-GMPpathway. Decreased levels of NO and increased levels of arginase (which degrades a precursor molecule in the NOS pathway) have been reported in PE (54-55). A deficiency in NO has been shown to correlate with met abolic derangements seen in PE, such as hypertension, proteinuria, and platelet dysfunction. NO deficiency induces the uteroplacental changes characteristic of PE in pregnant mice, including decreased uterine artery diameter, spiral artery length, and uteroplacental blood flow. These findings suggest that an intact NOS system is essential for normal spiral artery remodeling and pregnancy.

Oxidative Stress

From the start of pregnancy, the placenta assumes a state of oxidative stress arising from increased placental mitochondrial activity and production of reactive oxygen species, mainly superoxide anion. In PE, a heightened level of oxidative stress is encountered (56) from the placenta, when free radical synthesis occurs with maternal leukocytes. The superoxide–producing enzyme NADPH oxidase is present in placental trophoblast and women with early onset of PE have been found to have higher superoxide production compared with those with late-on set disease (57).

Sterile Inflammation and PE

Sterile inflammation is triggered when Damage associated molecular patterns (DAMPs) activate Pattern recognition receptors (PRRs) to mount an acute immune response in order to solve the adverse triggering insult. Since DAMPs are endogenous intracellular molecules primarily released as a result of non-programmed cell death to convey danger cues in the first few hours of an injury, they are also referred to as alarm ins. Candidate alarmins include, but are not limited to: high mobility group box1(HMGB1), Decorin, von Willebr and factor(VWF), Heat shock proteins (HSP) and uric acid. Placental hypoxia elicitsa variety of cellular and inflammatory responses in PE, elevating circulating levels of many pro-inflammatory factors (Figure 3).

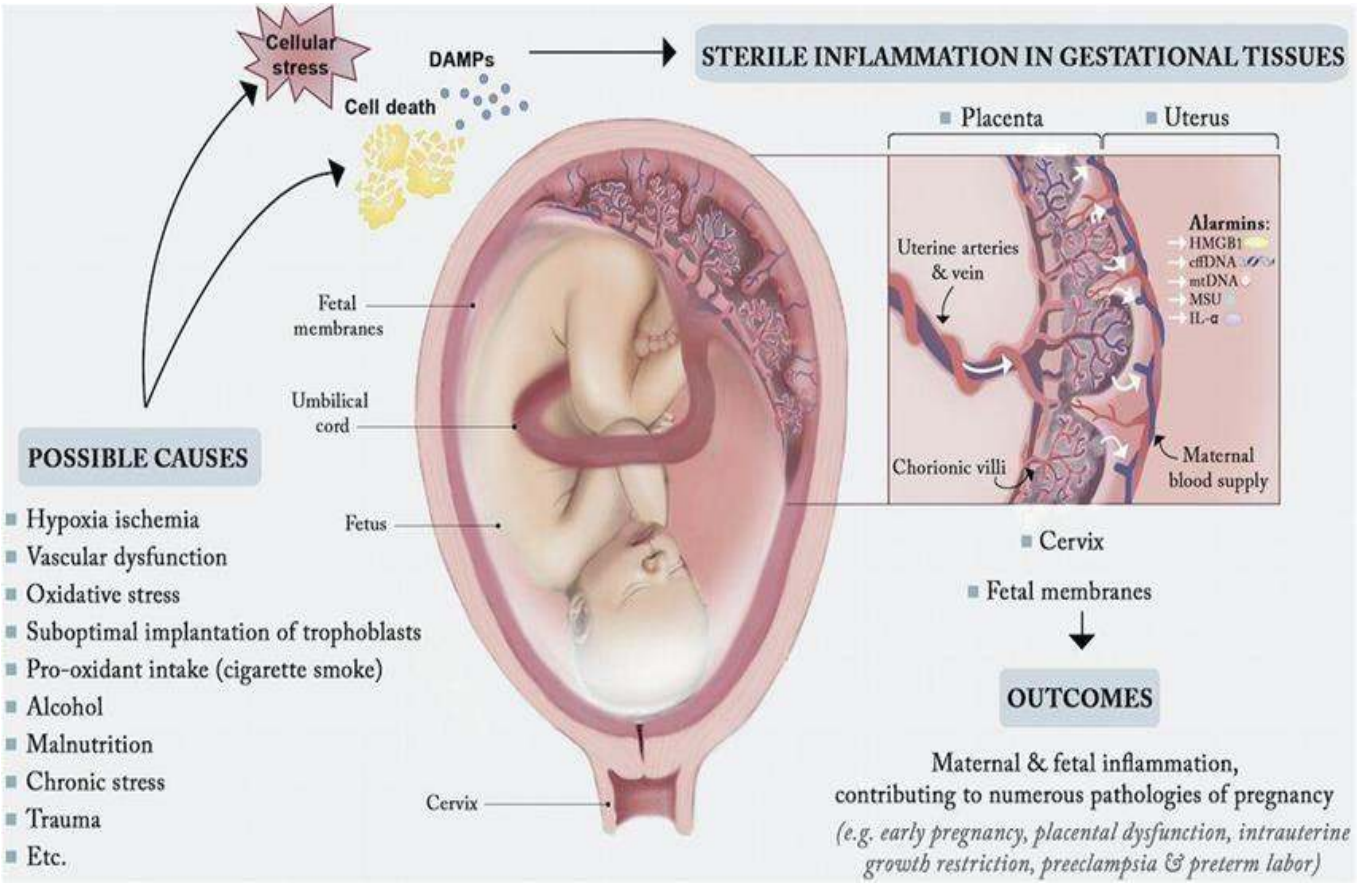

Figure3: Sterile inflammation and PE.

HMGB1

HMGB1 is a highly conserved non-histone protein (25 kDa) with cytokine-like activity in the extra cellular space. It is ubiquitously expressed in the nucleus where it plays a role in DNA replication, transcription and repair, and nucleo some stabilization. Although originally discovered in nucleus, HMGB1 is also found in cytosol, mitochondria and on membrane surface, and can be released to the extracellular milieu through active (secretion) and passive pathways: (I) active pathways are triggered by pathogenic products (e.g. bacteria, viruses) or other stressors (e.g., oxidative stress, cytokines), which has been shown in immune cells and non-immune cells; where as (ii) passive release is observed following tissue injury and cell death, especially necrosis and in specific cases of apoptosis–including when triggered by sterile injury events (e.g., hypoxia, senescence, auto immune disease).

HMGB1 fundamentally resides in the nucleus under normal circumstances, stabilizing nucleo somes and facilitating gene transcription (58). However, under different conditions, it is hyperacetylated, translocated from the nucleus to the cytosol, and then secreted actively or passively (50). It has been shown that HMGB1 passively diffuses from various cells to the extracellular space during cellular necrosis or damage (60). It is also actively released by macrophages, monocytes, and dendritic cells upon activation (61). Once HMGB1 accumulates in the extracellular milieu, it conveys danger signals by triggering inflammatory pathways via Toll-like receptors. A study showed that the mechanism of HMGB1-induced inflammation is mainly mediated via the MyD88-dependent pathway(62).

HMGB1causes endothelial dysfunction (63) and promotes leukocyte activation and thrombosis (64). Increased HMGB1 expression has been reported in the placenta or placental explant-derived extra cellular vesicles from PE patients (65-66). Hypoxic trophoblastic cells in case of PE are rich sources of numerous alarmins such as uric acid, cell-free fetal DNA, HMGB1, and IL-1α; when they die, they release these alarmins into the extracellular environment which can result in a sterile inflammatory response (67). HMGB1 is expressed by trophoblasts and can be found either in their nucleus or cytoplasm (68). When the placentae are exposed to preeclamptic blood serum of the mother, there is increase in levels of cytoplasmic HMGB1 in the syncytiotropho blast (69). Praderv and et al. showed that circulating levels of HMGB1 are higher in third-trimester PE than in normal pregnancies (70). This evidence suggests that HMGB1 plays a role in the development of PE.

Decorin

Decorin is a small leucine-rich proteoglycan produced by connective tissue and highly expressed in reproductive tissues such as chorionic villusmesenchymal cells and decidual cells of the pregnant endometrium. Biological functions of DCN include: collagen assembly, myogenesis, tissue repair and regulation of cell adhesion and migration by binding to ECM molecules or antagonizing multiple tyrosine kinase receptors (TKR) including EGFR, and VEGFR-2(71). DCN restrains angiogenesis by binding to thrombospond in-1,TGF β and VEGFR-2. DCN actions at the fetal-maternal interface include restraint of trophoblast migration, invasion and uterine angiogenesis. DCN over expression in the decidua is associated with PE and may have a causal role by compromising endovascular differentiation of the trophoblast and uterine angiogenesis, resulting in poor arterial remodeling. Elevated DCN level in the maternal blood has been suggested as a potential biomarker in PE (72).

In a study, Gogiel *et al*. reported increased DCN levels of the umbilical cord vein wall inpatients with PE (73) and Siddiqui et al. found that increased DCN levels were predictors of PE even before the on set of clinical symptoms (74). The link between DCN and PE can be attributed to the impaired proliferation and migration of trophoblasts and endothelial dysfunction, which are thought to be responsible for adverse pregnancy outcomes.

vWF

Von Wille brand factor(vWF), a multi-domain multi merized glycoprotein, plays an important role in primary hemostasis. vWF is a carrier protein for blood clotting factor VIII (FVIII) and this interaction is required for normal FVIII survival in the circulation. Under physiological conditions, the vascular endothelium produces many substances which are closely associated with hemostasis, fibrinolysis, the synthesis of growth factors, and the regulation of vessel tone and permeability. One such substance that is synthesized by, and stored in,endothelial cells is vWF. vWF is also known to have an important function in plate let adhesion an daggregation. However, vWF is as ensitive marker and levels can be influenced by many pathological conditions, including the acute phase response (75).

A study showed that VWF levels are elevated in healthy pregnancy compared with the non pregnant state, and further increases in pregnancies complicated by PE and that pregnancies complicated by severe PE exhibit VWF biology that is distinct from healthy pregnancy(76). This speculates that PE-associated VWF differences could play a direct role in the vascular pathogenesis and end-organ damage of PE.

HSP90

In pregnancies complicated with PE, diminished placental function increases the apoptosis in placenta endothelial cells, where an increase of HSP90 protein levels has been reported when compared to that observed in normotensive controls. HSP 90, the most abundant chaperone in eukaryotic cells accounting for 1–2% of cell proteins in most tissues, is involved in multiple biological processes such as cellproliferation, differentiation, and apoptosis.

Uric acid

Uric acid (160 Da) is a product of the metabolic breakdown of purine nucleotides by xanthine oxidase, with normal blood concentration range between 40-60 μg/ml. Upon achieving concentrations >70 μg/ml, uric acid forms needle-like, immunostimulatory monosodium urate (MSU) crystals, which cause acute inflammation. In the last few years, uric acid has been vastly regarded as an alarmin of sterile inflammation because of the high cytosolic concentration (≈ 4mg/ml) released upon cell death, which reacts with extracellular sodium to form MSU in the immediate vicinity of cellular injury.

CNAPS

Circulating nucleic acids in maternal plasma and serum (CNAPS) is a group of extra cellular nucleic acids including cell free DNA (cfDNA), cffDNA (fetal origin), total circulatingRNA, placental RNA, and other novel species.

Multiple pathways have been associated with the pathophysiology of PE, including inadequate placental development and systemic maternal endothelial dysfunction. As a link between impaired placentation and endothelial function, both maternal and fetal-derived cell-free deoxyribonucleic acid (cfDNA) fragments have been found in the circulation of pregnant women (77), and are known to be elevated in PE compared to normal pregnancy (78-79). In addition, increased cfDNA levels in maternal circulation correlate with disease severity in PE, and are associated with poor maternal-fetal outcomes (80-81).

Cellular damage and/or tissue-associated hypoxia leads to elevated RNA fragments, extracellular RNA (eRNA), in the circulation released from the disrupted, damaged cells (82). Asper background literature survey, we found that eRNA initiates cascades related to vascular diseases (83-84) i.e., that of blood coagulation along with inflammatory processes (85). Identification of endogenous ligands (DAMPs) by contemporary studies has created a substantial interest due to their high significance in SI. Tissue hypoxia can stimulate apoptosis and necrosis of affected tissue resulting in liberation of extra cellular nucleic acids in to circulation.

The exact underlying mechanism leading to the release of extracellular DNA into the maternal circulation and its consequencesare not completely clear, butexisting evidence indicates that fetal cell trafficking is perturbed in placental tissue early in gestation, and predates the manifestation of clinical symptoms in PE (86). It has been hypo the sized that increased circulating levels of fetal cfDNA may be due to ischemia/hypoxia within the intervillous space resulting in enhanced oxidative stress, apoptosis and/or necrosis of trophoblastic cells (87). Recent studies suggest that fetal cfDNA may activate specific receptors in maternal immune cells and platelets, which in turn trigger the production and secretion of pro-inflammatory cytokines and additional cfDNA release, resulting in a systemic humoral and cellular immuneresponse in PE (88). Interestingly, cfDNA may also act on the maternal vasculature, possibly contributing to endothelial and vascular dysfunction and then hypertension in PE (89).

Munchel *et al*. examined circulating RNA in pregnant women who developed early-onset PE and found 49 transcripts of maternal, placental, and fetal origin that classified as mall but in dependent cohort of pregnant women with early-onset PE (90). Several studies have suggested that dysregulated non-coding RNAs in the maternal-fetal interface participate in the regulation of proliferation, invasion, and apoptosis of trophoblasts, thereby promoting the pathogenesis of PE(91-92).

CONCLUSION

PE is a major cause of maternal mortality & morbidity particularly in low- and middle-in come countries(LMICs). Multi-faceted factors including hypoxia, inflammation of undetermined mechanisms impact the on set & progression of this disease which cause immunological alteration of placental micro-environment & impaired angiogenesis. PE, characterized by hypertension and inflammation, is accompanied by multiple symptoms including proteinuria, inflammatory response and oxidative stress which have been implicated as the key features in the pathophysiology of PE. As previously reported, hypertension and proteinuria are the characteristic symptoms of PE which has also been proven in our clinical assessment.

One of the contributors of PE is placental hypoxia leading to oxidative stress. As NO is a potent vasodilator, it relaxes the smooth muscle contraction of blood vessels. When its level in circulation decreases, the contraction of blood vessels increases which leads to elevation in the blood pressure. As a result of hypoxia, NO levels decrease in the blood vessels causing hypertension in the PE patients.

Inflammation is strongly associated with placental dysfunction and complications related to pregnancy such as preterm birth (PTB), low birth weight, fetal growth restriction (FGR), still birth and PE. Our knowledge of the actions of DAMPS at the maternal- fetal interface is still in early stages. Some of the important DAMPs that play a vital role in pathophysiology of PE areHMGB1, DCN, vWF, HSP90, circulating nucleic acids, ands 100b. As an inflammatory cytokine, HMGB1 plays an important role in endothelial permeability under oxidative stress and promotes the pathological process of inflammation. Placental is chemia may promote the release of a variety of factors, such as vWF, into the maternal circulation synthesized by the endothelium. vWF has been suggested to be a marker of generalized endothelialdys function, which leads to the clinically recognized symptoms of PE, which include hypertension, proteinuria, thrombocytopenia and impaired

liver function. Decorin (DCN) is a protein that limits the invasion and endovascular differentiation of extravillous trophoblast cells during early placentation and is found in creasing quantities in maternal blood and placental tissue in PE patients. Apart from hypertension and proteinuria PE also manifests with liver and endothelium dysfunction (93). It is deduced that in PE there is release of different mediators from liver and blood vessel endothelium which causes vasoconstriction and liver hypoxia, leading to the diagnosis of systemic disorders such as neuronal edema, cardiovascular disorder, liver disorders etc.

In normal pregnancy, the utero-placental spiral arteries in the myometrium undergo structural modifications where the musculo-elastic tissue of the tunica media is replaced by invading trophoblastic cells surrounded by a thick layer of fibrinoid material. The affected vessels undergo progressive vasodilatation. This phenomenon is described as the *"physiological changes of spiral arteries"*. These changes are responsible for the low vascular resistance of the placental bed and allow a large increase in blood flow to the intervillous space. In pre-eclampsia, the endovascular trophoblast does not invade the myometrium. Physiological changes are confined to the decidual segment. Therefore, the spiral arteries in the placental bed are less dilated than normal pregnancy and they remain responsive to vasomotor influences. The impaired intervillous blood flow results in inadequate perfusion and ischemia of the placenta especially in the second half of pregnancy (94). The histological changes in PE/eclamptic placentas include infarcts, increased syncytial knots, hyper vascularity of the villi, cytotrophoblast proliferation, thickening of the trophoblastic membrane, obliterative enlarge dendothelial cells in the fetal capillaries, and atherosis of the spiral arteries in the placental bed (95).

Future research can be focused on large-scale genomic and proteomic approaches for the identifying of novel target molecules and diagnostic biomarkers, which will aid in formulating effective therapeutic interventions to prevent PE and/ordetect and treat it at an early stage.

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