Pre-eclampsia: pathophysiology and clinical implications

Graham J Burton, Christopher W Redman, James M Roberts, Ashley Moffett

[1]Department of Physiology, Development & Neuroscience, University of Cambridge, UK

[2]Centre for Trophoblast Research, University of Cambridge, UK

[3]Nuffield Department of Obstetrics and Gynaecology, University of Oxford, UK

[4]Magee-Womens Research Institute, Depts. Obstetric Gynecology and Reproductive Sciences, Epidemiology, and Clinical and Translational Research, University of Pittsburgh, USA

[5]Dept of Pathology, University of Cambridge, UK.

Correspondence to: A Moffett
am485@cam.ac.uk

Series explanation: State of the Art Reviews are commissioned on the basis of their relevance to academics and specialists in the US and internationally. For this reason they are written predominantly by US authors.

ABSTRACT

Pre-eclampsia is a common disorder that particularly affects first pregnancies. The clinical presentation is highly variable but hypertension and proteinuria are usually seen. These systemic signs arise from soluble factors released from the placenta as a result of a response to stress of syncytiotrophoblast. There are two sub-types: early and late onset pre-eclampsia, with others almost certainly yet to be identified. Early onset pre-eclampsia arises owing to defective placentation, whilst late onset pre-eclampsia may center around interactions between normal senescence of the placenta and a maternal genetic predisposition to cardiovascular and metabolic disease. The causes, placental and maternal, vary among individuals. Recent research has focused on placental-uterine interactions in early pregnancy. The aim now is to translate these findings into new ways to predict, prevent, and treat pre-eclampsia..

Introduction

Eclampsia has been documented for more than 2400 years, and features of the prodromal syndrome pre-eclampsia (previously referred to as toxaemia of pregnancy) have been documented for almost 200 years. The pathophysiology of these conditions, however, remains poorly understood, limiting therapeutic interventions. It has long been established that a placenta, but not a fetus, is required, and that the syndrome eventually resolves once the placenta is removed. Hence, in terms of pathogenesis it is primarily a placental disorder. Although commonly portrayed as a distinct entity, pre-eclampsia, at least its early onset variety, is just one in a spectrum of complications of pregnancy that share a common pathophysiology centered upon disordered placentation. That spectrum, referred to as "disorders of placentation" or the "great obstetrical syndromes," includes late spontaneous miscarriage, abruptio placentae, fetal growth restriction (FGR), pre-term rupture of the membranes, and premature delivery.[1] The lack of spontaneous pre-clinical animal models for these conditions has limited our understanding, but the recent advances in "omics technologies"[2] and the derivation of organoid cultures of the endometrium[3] and placental trophoblast[4][5] create new opportunities for systematic research.

This review considers modifications to the definition of pre-eclampsia, and the epidemiology, prediction, treatment, and long term consequences of the syndrome. In terms of the pathophysiology, the review summarises emerging evidence that there are at least two sub-types: early and late onset pre-eclampsia, with others almost certainly yet to be identified.[6] Early onset pre-eclampsia is widely acknowledged to have primarily a placental cause, while late onset pre-eclampsia may center around interactions between senescence of the placenta and a maternal genetic predisposition to cardiovascular and metabolic disease. While often presented as distinct sub-types, in reality the balance between the placental and maternal causations most likely varies among individuals across the spectrum of gestational age at clinical presentation. We discuss the pathophysiology in the light of recent advances of our understanding of the maternal-fetal interactions that take place in the first weeks following implantation, and emphasize the importance of the endometrium during the pre- and peri-conceptional periods for pregnancy success.

Sources and selection criteria

A PubMed search (pre-eclampsia OR preeclampsia) AND (placenta OR placental)] at the end of 2018 with no date restrictions yielded 10 611 citations. About 10% of our references date from the last century, the earliest to 1953. The rest were published in this millennium, the most recent in 2019, and more than one third in the last five years. We considered only articles published in English. Additional articles and references were obtained by searching the bibliography of published papers. Most of the chosen references are exclusively data based. Trivial, repetitive, or inconsistent research reports were rejected. In this way, we summarized what is known about the pathophysiology and clinical aspects of pre-eclampsia.

Definition

Defining pre-eclampsia is difficult because it is a syndrome characterized by a group of clinical features that, when they occur together, lead to diagnosis and treatment. There is no gold standard, and all the features

are, in isolation, non-specific. Numerical features such as arterial blood pressure or proteinuria are defined by thresholds, which themselves are arbitrary. Hence, while the definitions seem precise, they are not securely based and leave important uncertainties. Only a definition based on unique pathogenic feature(s) will resolve this unsatisfactory situation.

Until recently, the accepted definition of pre-eclampsia was new onset hypertension and proteinuria developing in the second half of pregnancy and resolving after delivery. The more common and less dangerous new onset hypertension without proteinuria was called gestational hypertension.[7] Subsequently, refinements have been proposed, but new onset hypertension remains common to all versions. Currently, the diagnosis endorsed by the International Society for the Study of Hypertension in Pregnancy (ISSHP) embraces new onset hypertension (systolic >140 mmHg and diastolic >90 mmHg) accompanied by one or more other features: proteinuria, other maternal organ dysfunction (including liver, kidney, neurological), or hematological involvement, and/or uteroplacental dysfunction, such as fetal growth restriction and/or abnormal Doppler ultrasound findings of uteroplacental blood flow.[8] The categories of hypertension in pregnancy recognised by the ISSHP are shown in fig 1.

As the pathophysiology becomes clearer, assays of biochemical markers, such as maternal concentrations of angiogenic or anti-angiogenic factors, are being developed to improve diagnosis and prediction.[9 10] These assays may eventually be incorporated into more precise definitions of pre-eclampsia, and of related placental syndromes. Consequently, hypertension itself is not a necessary part of the syndrome.[11]

Both the ISSHP and the American College of Obstetricians and Gynaecologists recommend that the terms "severe" and "mild" pre-eclampsia should no longer be used, as all cases are potentially threatening clinically.[8] By contrast, the distinction between the early and late onset forms of the syndrome is increasingly recognized, with a watershed of 34 weeks' gestational age.[6]

Epidemiology

The complexities of defining pre-eclampsia affect the accuracy of determining its incidence, especially across different countries. Ascertainment is incomplete in low and/or middle income countries (LMIC), and standardization of diagnostic accuracy is almost impossible. Rates based on institutions will be biased by referral of cases, especially in tertiary centers where most research is con-
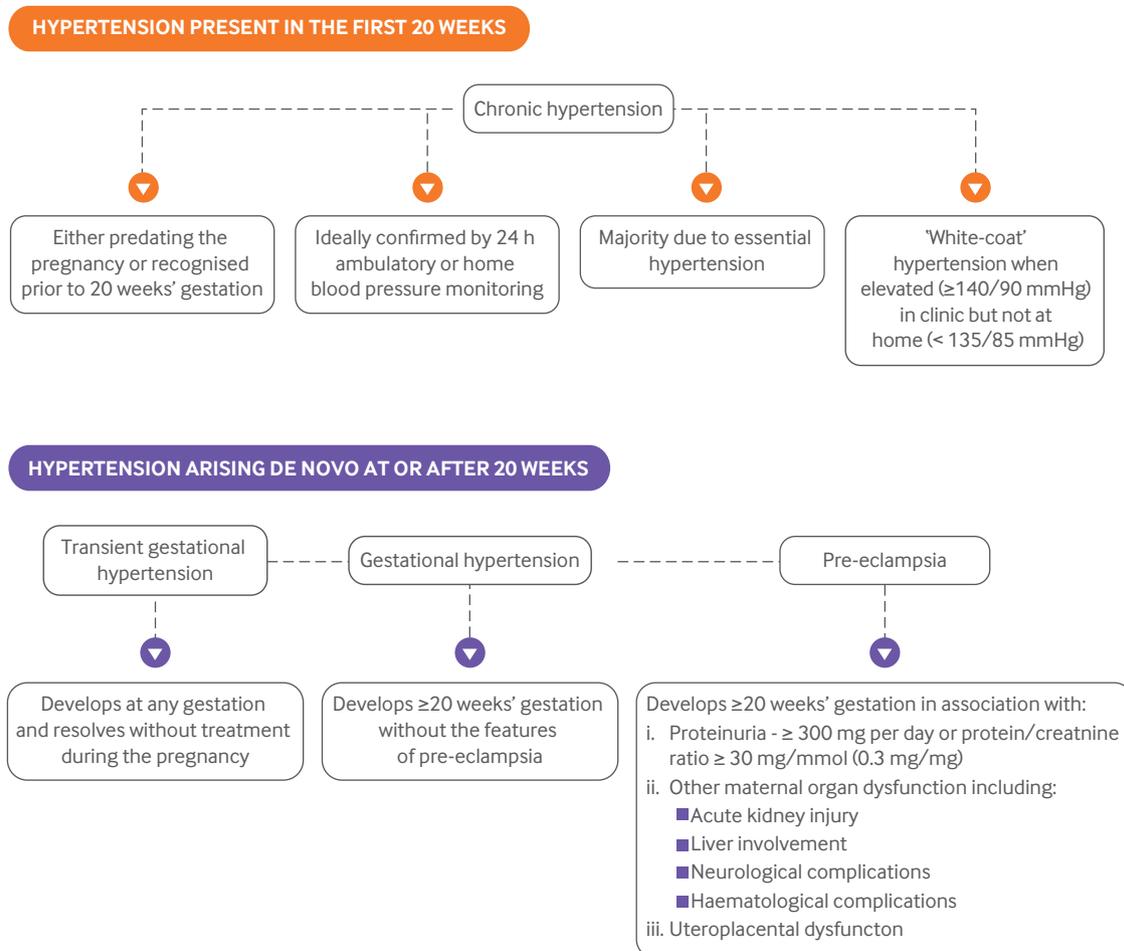
HYPERTENSION PRESENT IN THE FIRST 20 WEEKS

Chronic hypertension

- Either predating the pregnancy or recognised prior to 20 weeks' gestation
- Ideally confirmed by 24 h ambulatory or home blood pressure monitoring
- Majority due to essential hypertension
- 'White-coat' hypertension when elevated (≥140/90 mmHg) in clinic but not at home (< 135/85 mmHg)

HYPERTENSION ARISING DE NOVO AT OR AFTER 20 WEEKS

Transient gestational hypertension

Gestational hypertension

Pre-eclampsia

- Develops at any gestation and resolves without treatment during the pregnancy
- Develops ≥20 weeks' gestation without the features of pre-eclampsia
- Develops ≥20 weeks' gestation in association with:
  i. Proteinuria - ≥ 300 mg per day or protein/creatnine ratio ≥ 30 mg/mmol (0.3 mg/mg)
  ii. Other maternal organ dysfunction including:
    ▪ Acute kidney injury
    ▪ Liver involvement
    ▪ Neurological complications
    ▪ Haematological complications
  iii. Uteroplacental dysfuncton

Fig 1 | Categories of hypertension in pregnancy recognised by the ISSHP[8]

ducted. A global estimate derived from data of nearly 39 million pregnancies suggests an incidence of 4.6%.[12] Wide regional differences are found, with a reported incidence as low as 0.4% in Vietnam.[13] The condition is especially common in women indigenous to, or with ancestry from, sub-Saharan Africa.[14] The incidence of eclampsia is lower but quite variable, ranging from 0.015% in Finland[15] to an estimated 2.9% in some parts of Africa,[12] illustrating that the rate depends in part on access to obstetric care.[16]

Maternal mortality from pre-eclampsia/eclampsia is highest in LMIC, and worldwide accounts for at least 63 000 maternal deaths per annum. In high income countries, most progress in reducing the maternal toll was made during the period 1940-1970.[201] Over the same time, major breakthroughs in clinical management were few, and the substantial improvements in maternal death rates from pre-eclampsia/eclampsia were achieved by empirical advances in care, professional education, higher clinical competence, and, more recently, consistent application of national guidelines such as in the UK from the National Institute for Health and Care Excellence (NICE).[17] In LMIC, which lack equivalent resources, pre-eclampsia accounts for nearly 30% of all maternal deaths in 29 countries (20 per 100 000), a mortality rate of 0.8% for affected women.[202] This is more than 200 times higher than the mortality specific rate of 0.03% in the UK, assuming that the national incidence of pre-eclampsia is about 3%.[203]

Risk factors listed in box 1 represent data from three systematic reviews.[13 18 19] However, as there are likely multiple pathophysiological sub-types, it cannot be expected that all risk factors will be shared. The risk of pre-eclampsia is higher in a first pregnancy (~4%), and there is a protective effect of a normal first pregnancy with lower risk (~2%) in subsequent pregnancies. The risk of recurrence is high; ~15% after one pre-eclamptic pregnancy and ~32% after two pregnancies in a cohort of nearly 800 000 pregnancies in Sweden, with some confounding effect from a longer interbirth interval.[20]

Fetal sex is increasingly recognised as an important risk feature. A meta-analysis based on 219 575 singleton pregnancies, of which 9033 developed pre-eclampsia, found that when the pregnancy delivered at term (≥37 weeks) there was no difference in the sex ratio.[21] However, a predominance of female fetuses was found in those pregnancies delivering before 34 weeks (odds ratio 1.36, 95% confidence interval 1.17-1.59). Analyses of sex differences in placental gene expression indicate that almost half are X linked and arise from escape of X inactivation.[22 23] Thus, the male fetus may be more susceptible to suboptimal placentation, or less adaptable to adverse conditions.[24 25] This may reflect sex differences in uteroplacental malperfusion. The uterine artery pulsatility index is higher, and notching of the Doppler waveform more common in women carrying a male compared with a female fetus, indicating greater vascular resistance.[26] Hence, early loss of more severely impaired pregnancies carrying male fetuses could explain the female sex bias of early onset pre-eclampsia.[21]

The primary role of the placenta

Factors emanating from the placenta into the systemic circulation are considered to result in the maternal syndrome of pre-eclampsia.[27] Oxidative stress of the syncytiotrophoblast, the cell type that forms the epithelial covering of the placental villi in contact with maternal blood, is one of the hallmarks, particularly in the early onset form.[28 29] When stressed, the syncytiotrophoblast releases a complex mix of factors, including pro-inflammatory cytokines, exosomes, anti-angiogenic agents, and cell-free fetal DNA, into the maternal circulation. These disrupt maternal endothelial function resulting in a systemic inflammatory response, the clinical syndrome of pre-eclampsia (fig 2).[27 30] Different stressors can perturb the syncytiotrophoblast, but the main one in early onset pre-eclampsia is uteroplacental malperfusion secondary to defective remodeling of the uterine spiral arteries.[1] By contrast, in late onset cases the cause is more likely an increasing mismatch between normal maternal perfusion and the metabolic demands of the placenta and fetus, coupled with a maternal predisposition to inflammation, a high BMI, and/or a high arterial pressure.[31] To understand the primary underlying defect in the spectrum of placentally related complications of pregnancy, it is necessary to focus on early events in the development of the placenta.

Placental development, spiral artery remodeling, and early onset pre-eclampsia

Placental development is precocious. By the end of the third week post-fertilization a shell of trophoblast cells encapsulates the conceptus and forms the interface with the maternal tissues. It is essential that a robust shell is formed, as it seals off the conceptus and protects it from excessive levels of oxygen and xenobiotics during the critical phase of organogenesis.[32] The stimulus for this development is the histotroph or "uterine milk" derived from the endometrial glands. These secretions, which are rich in nutrients and mitogenic growth factors, are deliv-

---

Box 1 | Risk factors for pre-eclampsia from three systematic reviews[13 18 19]

- Chronic hypertension
- Antiphospholipid antibody syndrome
- Systemic lupus erythematosus
- Pre-gestational diabetes
- Chronic renal disease
- Multifetal pregnancy
- Pre-pregnancy BMI >30
- Previous stillbirth
- Nulliparity
- Maternal age >40
- Increased pre-pregnancy BMI
- Long inter-pregnancy interval (>5 years)
- Reduced school education
- Previous pre-eclampsia
- Assisted reproduction
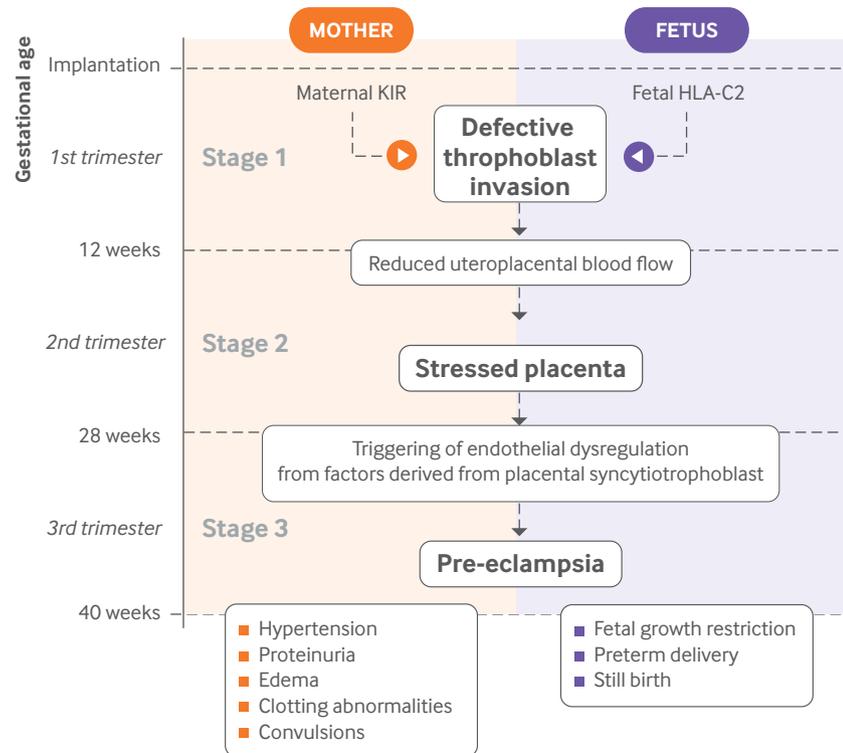- Previous intrauterine growth restriction
- Previous placental abruption

Fig 2 | Pathogenesis of pre-eclampsia with the subsequent effects on mother and fetus. The failure of trophoblast uterine interactions in the first trimester leads to a stress response in the placenta. This may affect growth and development of the villous tree, affecting transfer of oxygen and nutrients to the fetus. The stress to the syncytiotrophoblast leads to shedding of a range of factors into the systemic circulation. These factors cause a systemic inflammatory response resulting from disruption of the homoeostatic functions of the maternal endothelium, including regulation of clotting, fluid transfer, and blood pressure

ered into the placenta where they bathe the early villi, which contain the progenitor trophoblast cells.[33] Suppression of gland development in sheep and mice shows that the secretions are essential for early placental development.[34] Growth factor expression in the glands is up regulated during early pregnancy in response to lactogenic hormones derived from the trophoblast. The placenta is therefore able to stimulate its own development through a dialogue involving the decidua and the endometrial glands. We speculate that the same dialogue operates in humans, although there are no firm data at present.[35] It is possible to address this question directly now that organoid cultures of the endometrial glands[3] and of the trophoblast[4 5] can be generated. Co-culture experiments will enable identification of the impact of trophoblast signals on the gland secretome, and assessment of the impact of that secretome on trophoblast proliferation and differentiation.

Failure of that dialogue could cause incomplete development of the cytotrophoblast shell and lay the pathophysiological foundations for most placentally related complications. When severe, deficient formation of the shell is associated with spontaneous miscarriage, even in karyotypically normal pregnancies.[36] It seems likely that, in less severe cases compatible with an ongoing pregnancy, it predisposes to pre-eclampsia, as the shell is also the source of the extravillous trophoblast cells (EVT)

which are required for remodeling the maternal spiral arteries that ultimately supply the placenta. Evidence to support this hypothesis is lacking, but transcriptional profiling reveals defects in decidualization in women who either develop or have experienced pre-eclampsia.[37 38] In addition, the risk of pre-eclampsia is increased following two miscarriages,[39] pointing to defective interactions between trophoblast and the decidua.

The EVT arise from the outer surface of the shell through a partial epithelial mesenchymal transition, transforming into invasive cells characterized by expression of HLA-G. The cues and signalling pathways regulating this transition are not known, but the recent ability to derive EVT from trophoblast stem cells[40] and from trophoblast organoid cultures[5] will provide a powerful tool in understanding the process. Individual EVT migrate through the actions of matrix metalloproteinases via two routes. Initially, interstitial EVT migrate through the stroma towards the spiral arteries. These are the terminal branches of the uterine vasculature within the endometrium that ultimately supply the placenta. In the non-pregnant state, the arterial walls contain extensive smooth muscle that is highly responsive to endocrine and vasoactive stimuli. During normal pregnancy, EVT destroy the smooth muscle and elastin, which are replaced by inert fibrinoid material.[41] Although the mechanisms underpinning remodeling are not fully

understood,[42] the presence of EVT around the artery is essential. Endovascular trophoblasts then pass down the lumens of the spiral arteries, forming aggregates of cells that effectively plug the arteries during the first weeks of gestation.[43] Eventually, interstitial EVT move through the stroma to reach the inner third of the myometrium where they fuse to generate static, multinucleated giant cells.

Many studies have focused on control of this invasion,[44] and have largely used reductionist systems in vitro involving choriocarcinoma and other trophoblast-like cell lines. Extrapolating the results in vivo is difficult as local interactions that occur with multiple maternal cells, cytokines, and glycoproteins will be critical. Single cell RNA sequencing of the maternal and fetal cells present in the placental bed during the first trimester has predicted several potential receptor-ligand-receptor interactions.[45] Reports on the extent of the invasion in pre-eclampsia have been conflicting. Some describe it as superficial,[46] but others found that it extends as deep as normal, but that the EVT fail to destroy the arterial walls.[47] This confusion reflects the difficulty in sampling all the spiral arteries in the placental bed in early human pregnancy.

Remodeling has two principal consequences for uteroplacental blood flow. Firstly, the terminal segments of the arteries dilate in a funnel shape as they approach the placenta. Remodeling in itself has a relatively minor impact on the volume of blood flow to the placenta, and hence oxygenation. By contrast, mathematical modeling suggests it reduces the velocity and pulsatility of the inflowing maternal blood by an order of magnitude to approximately 10 cm.s^{-1} (fig 3).[48] This reduction is essential to prevent damage to the delicate placental villi and microvilli, as can happen during placental perfusion in vitro if the flow rate is too high.[49] Secondly, trophoblast transformation of spiral arteries normally extends as far as the inner third of the myometrium. Hence, it includes the hypercontractile segment of the artery located in the junctional zone between the endometrium and myometrium that restricts blood loss during menstruation. This segment must be remodeled to prevent compromise of placental blood flow, while the more proximal elements of the uteroplacental vasculature dilate under other stimuli.

Recognizing that pre-eclampsia was associated with defective spiral artery remodeling during early pregnancy was a major step forward in the understanding of its pathophysiology.[50 51] However, subsequent histological studies of placental bed biopsies revealed that deficient arterial remodeling is not specific to pre-eclampsia, being common to the other disorders of placentation.[1] Indeed, non-transformed arteries can be seen in normal pregnancies.[52] Nonetheless, there is general agreement that maternal vascular lesions are more severe in cases of pre-eclampsia than in cases of fetal growth restriction alone, which in turn are more severe than in normotensive pregnancies.[47 52 53 54]

A further arterial lesion, acute atherosis, is seen at the end of gestation in the most severe cases. In a retrospective examination of 16 345 placentas, the lesion was found in 0.4% of uncomplicated pregnancies, compared with 10.2% of pre-eclamptic cases, most commonly the early onset form associated with growth restricted neonates.[55] The lesion is characterized by fibrinoid necrosis and accumulation of lipid laden intimal macrophages. Acute atherosis is not restricted to the placental bed and can affect any decidual non-transformed arteries. The location of the lesions suggests they arise because of the altered haemodynamics occurring when remodeling is deficient.[48] Unlike defective remodeling in itself, acute atherosis can severely restrict the caliber of the uteroplacental vessels, exacerbated by secondary thrombotic lesions, limiting the volume of blood entering the placenta and causing infarction with a risk of fetal demise. This effect has been seen using magnetic resonance imaging (MRI) in cases of early but not late onset pre-eclampsia.[56]

Deficient remodeling of the spiral arteries is often associated with high resistance uterine artery Doppler waveforms, leading to the assumption that one causes the other, although the physics of waveform determinants is complex.[57] In addition, challenging this assumption is the observation that identical changes in uterine artery waveform are seen during both normal and extra-uterine pregnancies where there can be no spiral artery remodeling.[58] Often overlooked is that the more proximal sections of the uterine vasculature: the radial, arcuate, and uterine arteries, undergo considerable dilation independent of trophoblast invasion. These effects are mediated through other factors, such as oestrogen and placental growth factor. Measurements taken from micro-computed tomography scans in mice show that the radial arteries account for ~90% of total uteroplacental vascular resistance,[59] and computational modeling indicates the same is true for humans.[60] Hence, more attention should be paid to more proximal elements of the uteroplacental vascular tree in future.[61]

Placental changes in pre-eclampsia

The gross placental lesions in pre-eclampsia principally reflect maternal malperfusion, with infarcts of the villous tissue at different stages of resolution, villous-free placental lakes, fibrin deposition, and inflammation. These lesions are not specific to the syndrome, but a meta-analysis found them to be four- to seven times more common in pre-eclamptic pregnancies than in normotensive controls,[61-63] with a spectrum of pathology that is more severe in early compared with late onset disease[64-68] (table 1).

At the microscopical level, there is focal necrosis of the syncytiotrophoblast with loss and distortion of the microvilli, dilation of the endoplasmic reticulum cisternae, and swelling of the mitochondria.[101 102] Hyperplasia of the underlying cytotrophoblast cells may be present, but some cells undergo degeneration or apoptosis.[101 103] Not surprisingly, these lesions are associated with shedding of trophoblastic debris.[96]

These morphological differences are backed by reports showing higher levels of placental stress at the molecular level. Thus, oxidative stress and activation of the unfolded protein response (UPR) are greater in early than in late onset pre-eclampsia.[94] One consequence of activation of the UPR is the suppression of non-essential protein
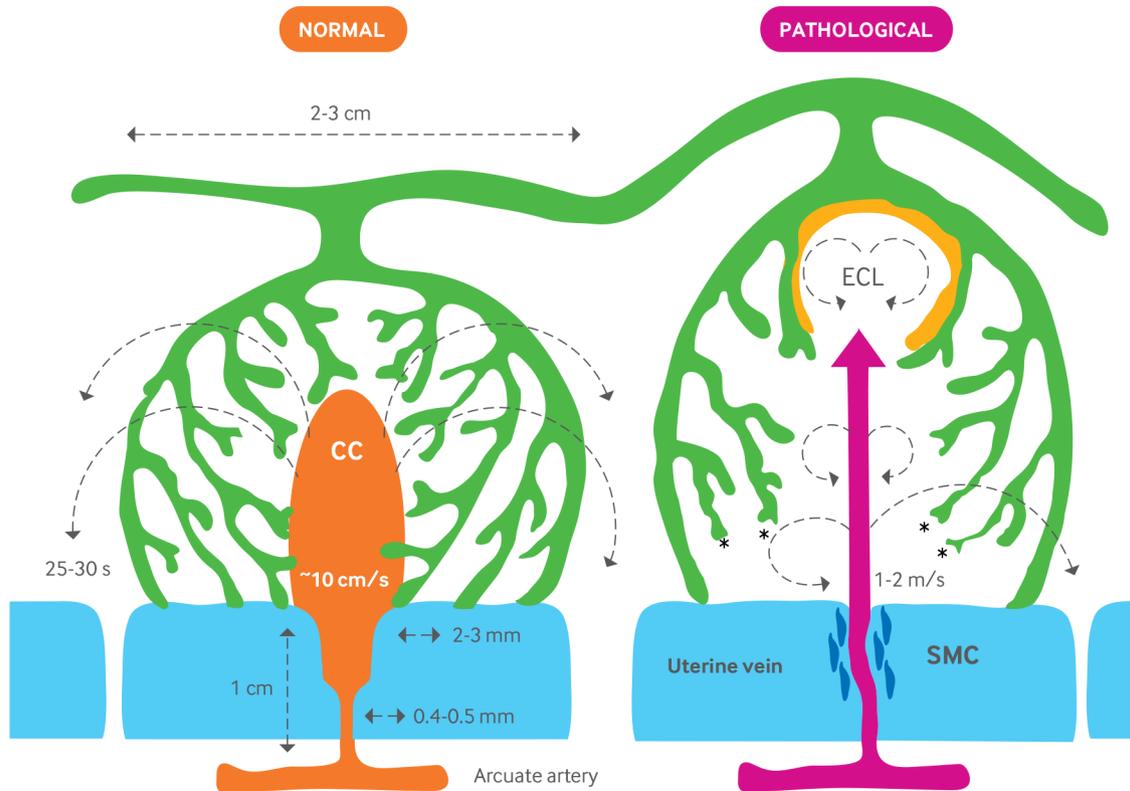
Fig 3 | Diagrammatic representation of the effects of spiral artery remodeling on the inflow of maternal blood into the intervillous space in normal and pathological pregnancies. Dilation of the distal segment of the spiral artery in normal pregnancies reduces the velocity of incoming blood, and the residual momentum carries the blood into the central cavity (CC) of a placental lobule, from where it disperses evenly between the villi. Transit time to the uterine vein is estimated to be in the order of 25-30 s, allowing adequate time for oxygen exchange. In pathological pregnancies, where no or very limited conversion occurs, the maternal blood enters the intervillous space in a jet-like spurt at speeds of 1-2 m/s. Flow is likely to be turbulent, indicated by the circular arrows, and the high momentum damages the villi, forming echogenic cystic lesions (ECL) lined by thrombus (stippled). The transit time will be reduced, so that oxygen exchange is impaired. Trophoblastic microparticulate debris may be dislodged from the villous surface. The retention of smooth muscle cells (SMC) around the spiral artery will increase the risk of spontaneous vasoconstriction and ischemia-reperfusion injury (Modified from[48] with permission)

synthesis, explaining the growth restriction often associated with early onset pre-eclampsia.[104] Particles from syncytiotrophoblast surface are released into the maternal circulation as microvesicles or secreted from internal multivesicular bodies as exosomes (nanovesicles). Both carry complex cargoes, including microRNAs,[105] and are potential liquid biopsies of the syncytiotrophoblast.[106] Shedding of placental microparticles is greater in early onset pre-eclampsia than in the late onset form,[96] while levels of exosomes in maternal serum are increased in early onset but not in late onset pre-eclampsia compared with age matched controls.[83] Exosomes may become important biomarkers of placental stress in the near future.[107]

Pre-eclampsia is also associated with changes in placental DNA methylation[93] and gene expression.[98 100] However, caution is needed in interpreting data, because the changes are likely to reflect the pathophysiology rather than being causal. Oxidative stress and similar changes in gene expression can be induced, for example, by the rigors of labor when the placenta is subjected to intermittent perfusion.[108] Mode of delivery and sample processing have major impacts on the placental transcriptome, metabolome, and activation of stress response pathways, and represent important confounding variables.[109] Additional factors, such as maternal administration of glu-

cocorticoids and other therapeutics, and the sex of the placenta should also be taken into account,[24 110] but are seldom reported. A further difficulty is the lack of non-labored, healthy pre-term control placental samples, as caesarean sections are rarely performed in obstetrically normal pregnancies at gestational ages equivalent to those in early onset pre-eclampsia. Placentas from spontaneous pre-term deliveries display high levels of stress due to either the predisposing pathology or vaginal delivery,[111] and so are often unsuitable as controls.

From placental stress to the maternal syndrome
In the classic two-stage model, placental stress leads to dysfunction of maternal peripheral endothelial cells, a systemic inflammatory response,[112] and the clinical syndrome of pre-eclampsia. Blood flow to maternal organs is reduced, and physiological assessment indicates vasospasm, activation of the coagulation cascade, and reduced plasma volume before clinical disease.[113-115]

Numerous placental factors could trigger the systemic syndrome, but the absence of a spontaneous pre-clinical model makes it difficult to elucidate their importance. As an alternative, an informative comparison may be that between FGR alone and FGR associated with pre-eclamp-

sia, because FGR shares much the same pathophysiology of deficient spiral artery remodeling with early onset pre-eclampsia, though to a lesser degree.[53 104 116 117] Different levels of placental cell stress may distinguish between the two conditions,[118] because at high levels of activation, the UPR switches from principally homoeostatic to pro-apoptotic and pro-inflammatory pathways.[119] Hence, the higher levels of placental senescence,[120] maternal serum pro-inflammatory cytokines,[121] cell-free fetal DNA,[122] leptin,[123] placental apoptotic debris,[124] soluble receptor (sFLT) for vascular endothelial growth factor (VEGF),[125 126] and the lower levels of placental growth factor (PlGF)[127] reported in early onset pre-eclampsia compared with FGR alone may reflect the severity of the initiating maternal malperfusion. Differences in the maternal responses to these factors will obviously contribute to the clinical manifestations.

Of the potential mediators listed, the balance between sFLT and PlGF is of particular clinical importance.[71] The elevated levels of sFlt are thought to bind and reduce the bioavailability of VEGF to the maternal endothelial cells, impairing their endogenous production of nitric oxide and causing vasoconstriction. By itself, sFlt does not cause activation of human umbilical endothelial cells in vitro, but it does render them more sensitive to pro-inflammatory cytokines.[128] This synergistic effect may explain why pre-eclampsia has proved so hard to treat, as it is likely that the peripheral aspects of the syndrome are caused by a complex mix of factors rather than any one mediator alone.

Because of the involvement of the endothelium, pre-eclampsia is a global systemic syndrome affecting many organs including the central nervous system, kidney, liver, and the coagulation cascade to varying degrees in different women. Metabolic abnormalities, including dyslipidaemia, insulin resistance, and inflammatory markers, are also characteristic.[129 130 131] There are striking differences in the severity and rate of progression of the disorder. Pre-eclampsia can present as a mild disorder that progresses slowly or one that develops rapidly to a life threatening condition. Changes in the maternal liver, adrenal glands, heart, and brain are consistent with decreased organ perfusion, and many are identical to those found in hypovolemic shock. The histological changes in the kidney are characteristic: these are concentrated in the glomerulus with profound endothelial swelling and disruption of the basement membrane and podocytes.[132] Seen in no other form of hypertension and reminiscent of hemolytic uremic syndrome (a thrombotic microangiopathy) these changes indicate that pre-eclampsia is not simply an unmasking of a propensity to hypertension. Pre-eclampsia usually resolves shortly postpartum. If removal of the placenta is delayed, for example in cases of extra-uterine pregnancies, the risk may continue for weeks or months until the placenta is reabsorbed.[133]

Genetics

A family history of pre-eclampsia and risk of recurrence has long been recognized, especially in those with the early onset form, stimulating a long search for the genetic predisposition to the disorder. However, pregnant women who have a monozygotic twin show no concordance, pointing to the role of maternal-fetal gene interactions.[134] That paternal genes are important is seen from the change of partner effect, and the increased risk with fathers born of an affected pregnancy or who previously fathered a pre-eclamptic pregnancy with another woman. The influence of the mother dominates, however, with variance of heritability estimated as 35% maternal, 20% fetal, 13% to a couple effect, and the rest to other effects.[135 136]

Pregnancy involves an interaction between maternal and fetal genes, which may explain the lack of success in finding genes associated with pre-eclampsia from studying maternal genomes alone. Using a candidate gene approach, the focus has been on genes likely to be involved in the final systemic stage of the disorder—particularly genes affecting endothelial function (eg, renin-angiotensin system), the oxidative stress, and thrombophilia pathways. Family linkage studies have also met with equally limited success. More recently, genome wide association studies (GWAS) have been performed. As with the candidate gene studies, these GWAS have been hampered by small numbers, lack of reproducibility, different ethnicities of participants, and problems with rigorous diagnostic criteria for pre-eclampsia. To date, no maternal sequence variants have been identified that can be replicated in independent datasets. However, one GWAS looking at fetal variants involving 4380 cases and 310 328 controls recently identified a variant near the gene encoding Fms-like tyrosine kinase 1 (FLT1), the receptor for vascular endothelial growth factor, in the fetal genome.[137] The association was strongest in cases of late onset pre-eclampsia, and when the birth weight exceeded the 10th centile. Thus, altered production of sFLT by the placenta in response to placental stress secondary to malperfusion may be affected by fetal genetic variants.

Since Medawar's seminal essay[138] pointing out that the fetus is "nature's transplant," the role of the maternal immune system in regulating successful pregnancy has been extensively studied. In addition, the key features of the immune system are memory and specificity, and pre-eclampsia occurs particularly in first pregnancies (memory) and after a change of father (specificity). Ideas in the field became dominated by the idea that immunosuppression is essential for successful pregnancy. A breakdown in maternal T cell tolerance was invoked as a cause of pregnancy disorders, but in humans multiple mechanisms exist to avoid decidual or systemic T cells killing placental trophoblast cells, and there is no evidence this is ever a cause of failing pregnancies.[139] Nonetheless, the immune system is programmed to discriminate between self and non-self, and thus uterine immune cells are likely to be able to detect the invading fetal trophoblast cells and potentially regulate the depth of invasion and transformation of the spiral arteries.

The main maternal leukocytes present at the time of implantation are not T cells but different types of lymphocytes, natural killer (NK) cells. Killer immunoglobulin-like receptors (KIR) are expressed by uterine NK cells (uNK)

Table 1 | Differences between early and late onset forms of pre-eclampsia

	Early onset	Late onset	System affected	Reference
Maternal serum				
Asymmetric dimethylarginine (ADMA)	↑	–	Vasoreactivity	69
Apelin	↑	–	Blood pressure, angiogenesis	70
sFlt/PlGF ratio	↑ ↑	↑/–	Angiogenesis, endothelial cell dysfunction	71 73
Brain natriuretic peptide	↑ ↑	↑	Cardiac dysfunction	74
Complement activation	↑	↑	Immune response	75
Copeptin	↑	–	Metabolic syndrome, insulin resistance	76
Fetuin-A	↓	↑	Inflammatory modulator, metabolism	77
Fibronectin	↑ ↑	↑	Clotting cascade	78
Growth differentiation factor-15 (GDF-15)	↓	↓ ↓	Cell injury, oxidative stress	79
HtrA1	↑	↓	Cell stress	80
Irisin	↓ ↓	↓	Adipomyokine, metabolism	81
Leptin	↑ ↑	↑	Metabolism	82
Placental exosomes	↑	↓	Placental stress	83
Progranulin	↑ ↑	↑	Cell growth	84
Pro-inflammatory cytokines	↑	–*	Inflammation	85
sVCAM-1	↑ ↑	↑	Endothelial dysfunction	78
R gulatory T cells	↓ ↓	↓	Inflammatory response	86
Maternal CVS function		▪		
Baroreceptor sensitivity	–	↑	Blood pressure	87
Heart rate variability	–	↑	Cardiac dysfunction	87
Left ventricular concentric hypertrophy	↑ ↑	↑	Cardiac dysfunction	74
Bilateral u erine artery notching	↑ ↑	↑	Uteroplacental blood flow	88
Placental perfusion	↓	↑	MRI placental blood flow	56
Placental				
Width	–	↑	Placental development	89
Pathological changes	↑	–*	Malperfusion	65 90
Villous volume, surface area	↓	–	Placental develpment	91
Total oxidant status	↑ ↑	↑	Oxidative stress	92
Altered methylation	↑	–	Oxidative stress	93
Placental stress	↑	–	Cell stress	94 95
Trophoblast debris	↑ ↑	↑	Maternal endothelial dysfunction	96
Mitochondrial copy number	↑	–	Metabolism	97
G-prote n coupled receptor signalling	↓	-	Immune and inflammatory responses	98
TLR4	↑	–	Immune regulation	99
EGFL7, ACVRL1	↓	–	Angiogenesis	100

*comparisons limited to early onset against late onset pre-eclampsia rather than age matched controls

and bind to HLA-C molecules on EVT. Because the maternal genes encoding KIR and the fetal genes encoding HLA-C are both highly polymorphic, there are different maternal KIR/fetal HLA-C genetic combinations in each pregnancy. Immunogenetic studies show that interactions of EVT with uNK cells are an important determinant of the risk of pre-eclampsia and other placentally related complications in both European and African populations.[140 141] From these, it appears that a degree of activation of the uNK cells is beneficial for successful pregnancy. uNK cells cluster around the spiral arteries, and it is presumed their activation causes the release of cytokines and proteases that stimulate the remodelling process, although evidence is still limited for how they function in normal and abnormal pregnancies.[142] This maternal/fetal genetic interaction system is only likely to account for a small amount of the genetic contribution to pre-eclampsia operating at the site of placentation. Other genetic influences will affect the stress response in the placenta (eg, sFLT1) and the maternal susceptibility to the systemic syndrome.

Screening

Pre-eclampsia is usually symptomless, making the syndrome hard to predict. Symptoms such as epigastric pain or severe headache frequently herald a terminal crisis, for example eclampsia or the HELLP (hemolysis, elevated liver enzymes, low platelets) syndrome, which requires prompt termination of pregnancy.[143] Screening of well women for the early stages of pre-eclampsia has been highly successful in limiting maternal and perinatal problems.[144] Antenatal care is based on predictions of the chances of pre-eclampsia developing before the next screening tests are due. Pre-eclampsia is uncommon before 20 weeks, but then progressively becomes more frequent towards term and beyond.[145] Hence, the frequency of checks is higher during the third trimester. Until recently, screening was based on timely detection of new onset hypertension and proteinuria, because they were the first features to be documented and their measurement is easy and cheap. They were introduced empirically and are effective overall. Confirmatory evidence comes from studies in LMIC, which consistently document the importance of adequate antenatal screening.[16 146]

In high income countries this situation has changed substantially. Impending or active pre-eclampsia can be detected by circulating biomarkers or Doppler ultrasound assessment of the uteroplacental circulation. This can be useful for the early onset but not late onset form of the

syndrome.[147 148] Circulating biomarkers may be placental or maternal. New hypertension or proteinuria are maternal markers of endothelial activation, whereas placental syncytiotrophoblast factors are further upstream in the pathophysiology and likely to be more precise. An increased ratio of sFlt-1/PlGF is a good marker of the placental component of pre-eclampsia, and of fetal growth restriction induced by placental malperfusion.[71]

The ability to exclude pre-eclampsia is also important, and before 35 weeks the PlGF value can rule out the need for delivery within the next two weeks with 98% probability.[149] When combined in the sFlt-1/PlGF ratio, this increases to a probability of more than 99% within the next week.[150] As with uterine artery Doppler assessment, the ratio does not predict late onset pre-eclampsia well. Ideally, diagnosis should be made early in pregnancy, when interventions could begin before the clinical features are manifest. Combinations of demographic and clinical factors with maternal blood pressure, uterine artery Doppler measurements, and blood biomarker assessments factors have been assembled to improve predictive efficiency.[31] A version of enhanced first trimester screening has been used in a trial of prophylactic low dose aspirin with encouraging results, identifying primarily early onset pre-eclampsia.[151] Whatever the outcome of these developments, measured rather than subjective estimates of the risk of pre-eclampsia should be robust.

Therapy
Prevention
Pharmacological and behavioural efforts to prevent pre-eclampsia are at best minimally effective. Evidence that a "healthy" diet,[152] "appropriate" weight gain and exercise,[153] and stress reduction[154] can reduce the risk is less than compelling; however, the general advantages of a healthy lifestyle justify these in pregnancy. Meta-analyses of the more than 40 000 women treated with aspirin in doses of less than 165 mg started in early pregnancy indicate a small beneficial effect to reduce the incidence of pre-eclampsia, fetal prematurity, and mortality.[155] Current recommendations for aspirin are for women with modest to high risk.[156] Calcium supplements of 1.5 to 2 g daily in settings with low calcium intake reduce the severity of blood pressure and adverse outcomes, and may reduce the incidence of pre-eclampsia or preterm birth.[157] The World Health Organization currently recommends this therapy for women in settings with low calcium intake.

The central role of placental oxidative stress in the pathophysiology provided a rationale for the administration of antioxidant vitamins. Clinical trials of antioxidant vitamins (C and E) have proved to be ineffective in several large interventional studies,[158] possibly because treatment was started between 10 and 20 weeks of gestation, after trophoblast transformation of spiral arteries. Alternatively, these vitamins may not be the appropriate antioxidants and others should be considered.[159 160 161] Whether therapy with vitamins or nutritional supplements before or in early pregnancy are effective[162 163] requires careful clinical testing. Preventive therapies for pre-eclampsia have been successful in small trials involving homogenous populations, but have not been supported by larger multicenter findings.[164]

Treatment of overt pre-eclampsia
Pre-eclampsia is a disease of two individuals, the mother and her fetus. Careful monitoring of the maternal condition and timely delivery with increasing progression of disease explain the difference in maternal mortality between HIC and LMIC, and the dramatic reduction of maternal mortality in HIC in the mid 20th century.[144] There were no deaths from pre-eclampsia in the latest Confidential Enquiry into Maternal Deaths in the UK. However, for the fetus, delivery may exchange a dangerous situation in utero for the morbidity and mortality of premature delivery. The maternal condition may be palliated to slightly prolong pregnancy and gain fetal maturation, but there is nothing currently that reverses the pathophysiology to improve outcome. Prolongation of the pregnancy increases the risk of fetal death in utero probably because of obstruction of uterine arteries by acute atherosis.

Palliation primarily involves antihypertensive therapy to avoid maternal intracranial bleeding, and magnesium sulphate as anticonvulsant therapy. The choice of antihypertensive agents, as determined by efficacy and safety for mother and fetus, is guided by a few trials but largely by experience.[130 165] Magnesium sulphate is more effective to treat or prevent seizures than other pharmacological agents,[166-168] and is safe if used appropriately. However, over-dosage results in respiratory and cardiac failure, and so its use should be monitored[169] and reserved for women where the risk-benefit ratio is acceptable.[130]

Long term implications
The consequences of pre-eclampsia for the mother and her baby are lifelong. Meta-analyses indicate a twofold increase in cardiovascular risk and death in women with previous pre-eclampsia.[170] This increases to six- to ninefold if pre-eclampsia occurs in more than one pregnancy, or before 37 weeks' gestation.[171] Specific risk increases are estimated as 3.7-fold for hypertension and 1.8-fold for stroke.[170] Less well studied, but related, is an increased risk of heart failure, particularly with preserved ejection fraction (HFpEF).[172] In addition, women with early onset pre-eclampsia can have diastolic dysfunction that persists in a subset for at least a year.[173] Microvascular disease with reduced capillary density is also more prevalent.[174]

An important question is whether similar risk factors cause pre-eclampsia and later life cardiovascular disease, or does pre-eclampsia cause later life cardiovascular disease? The answer requires information gathered before pregnancy. In the large HUNT study in Norway in which young women and men were recruited and followed, thus far for 20 years, 3225 women provided information before and after pregnancy.[175] Cardiovascular risk markers were compared before, and following, normal and pre-eclamptic pregnancies. Most, but not all, risk was present before the pre-eclamptic pregnancy. For example, women with pre-eclampsia have an increased prevalence of a gene variant known to be associated with

cardiomyopathy, and, indeed, 40% of women with peripartum cardiomyopathy have pre-eclampsia, both conditions commoner in women of African ancestry.[176 177]

The American Heart Association has emphasised that evaluation of cardiovascular risk for women should include a pregnancy history and that pre-eclampsia be considered a risk factor.[178] How this risk should be translated into follow-up is "work in progress."[179 180] Evidence based assessment of whether increased or earlier surveillance or pharmacological interventions are beneficial is currently underway.

An emerging area of some concern is the suggestion of increased cognitive dysfunction[181] and white matter lesions on MRI in the brains of formerly pre-eclamptic women.[182] Another intriguing relationship, but this time beneficial, is the reduced risk of breast cancer in women with a history of pre-eclampsia, which appears much greater if the fetus is male.[183]

The infant from a pre-eclamptic pregnancy also appears at increased risk for cardiovascular disease, although the relationship is not as well established as for the mother. Infants of pre-eclamptic mothers have higher blood pressure during young adulthood[184] and an increased risk for stroke in later life.[185] There are also disturbing reports of increased "mental disorders" including mood and anxiety in infants of women with pre-eclampsia, especially in females.[186] One third of infants of pre-eclamptic pregnancies are growth restricted and have the same increased risk of obesity, diabetes, hypertension, and other chronic diseases as other growth restricted infants.[187]

For the infant, the question is whether the associations are genetic and/or related to the post-delivery environment, or due to in utero exposure to the mother's pre-eclamptic pregnancy inducing epigenetic changes? At least one study suggests in utero exposure is not important, since adults exposed to pre-eclampsia in utero had the same cardiovascular risk as their siblings born from a non-pre-eclamptic pregnancy.[188]

Guidelines
Guidelines for the clinical management of the syndrome have recently been published by the ISSHP.[8]

Emerging therapies
There are several trials of drugs to alter the pathophysiology of pre-eclampsia. The statin pravastatin, administered from early pregnancy, is safe for mother and neonate.[189] Pre-eclampsia was less common in women receiving pravastatin (4 in placebo, 0 in cases n=10 in each group), although the trial was under powered.

A trial of the PDE5 inhibitor sildenafil citrate to prevent FGR found no prolongation of pregnancy,[190] with some adverse neonatal effects, including pulmonary hypertension, and should be discontinued.[191] Although a definitive benefit of the nitric oxide precursor arginine has not been established because of sample size and study design, potential benefit suggests further studies.[192 193]

An experimental approach is to reduce the concentration of sFlt in the maternal circulation using apheresis with a charge specific dextran sulfate column. In women presenting with very early onset pre-eclampsia, the pregnancy could be prolonged by eight and 15 days following single or multiple treatments, compared with three days in untreated controls.[194]

An alternative approach only tested in pre-clinical models is to knock down expression of sFlt in the placenta using interference RNA technology.[195] Other possible therapies are modifiers of endoplasmic reticulum stress, hydrogen sulphide donors,[161] and scavengers of fetal haemoglobin.[196]

Conclusion
Advances have been made in our understanding of the pathophysiology and clinical management of pre-eclampsia, but several research questions remain (box 2). The variability of clinical presentation, long range outcome, laboratory findings, and inconsistent response to preventive therapies and inaccuracy of prediction suggest different subtypes of pre-eclampsia.[164] As clinical phenotyping improves, complemented by analytical techniques[197] and data from large cohorts,[198] it is likely that the spectrum of pre-eclampsia will be better understood. At present, the early and late onset forms are the probable extremes of a spectrum. The early onset form is predominantly due to defective placentation during the first few weeks of pregnancy, and shares a common initiating pathophysiology to other disorders of placentation, especially FGR. Various strands of evidence indicate that the level of placental insult is greater in pre-eclampsia than in FGR, stimulating the release of a heavier burden of placental pro-inflammatory factors. The concentration of these factors, and their interactions with the maternal constitution, will determine the inflammatory response that distinguishes between the two conditions. By contrast, late onset pre-eclampsia appears to be driven by oxidative changes in the placenta induced by a progressive mismatch between maternal perfusion and feto-placental demands, coupled with a maternal predisposition to cardiovascular disease.

There are other current gaps in our knowledge. Racial disparities and differences in the frequency and severity of pre-eclampsia[199] are only partly explained by genetics

---

QUESTIONS FOR FUTURE RESEARCH

General
- What are the most accurate ways to predict pre-eclampsia?
- How many sub-types of pre-eclampsia exist?
- Does pre-eclampsia cause or exacerbate a predisposition to maternal cardiovascular disease?
- What impact will treatments designed to ameliorate the systemic syndrome have on fetal growth and survival?

Placenta related
- Can pre- or peri-conceptional interventions aimed at enhancing maternal wellbeing and endometrial function prevent pre-eclampsia?
- Why is pre-eclampsia seen particularly in first pregnancies?
- What is the role of paternally derived fetal genes in placental development?
- What are the similarities and differences in the pathophysiologies of other placental-related complications (eg, preterm labour, fetal growth restriction, unexplained stillbirth, and abruption) compared with pre-eclampsia?

and socioeconomic status. There is an increased risk of all disorders of placentation in women with recent African ancestry that remains unexplained. What are the other causal factors? Can we learn about the unique features of cardiovascular disease in women by understanding the pathophysiology of pre-eclampsia? What are the short and long term implications of pre-eclampsia for the offspring? Are the effects of pre-eclampsia due to more than prematurity or fetal growth restriction? These and other poorly understood features of pre-eclampsia suggest targets for future research.[200]

The substantial reduction in maternal and fetal mortality and morbidity that has been achieved over recent years has depended almost entirely on improved clinical awareness and care. To improve prediction and prevention it is now important to augment these advances with increased understanding of placental development, interactions with the uterine lining, and pathophysiology across the entire duration of pregnancy.

1 Brosens I, Pijnenborg R, Vercruysse L, Romero R. The "Great Obstetrical Syndromes" are associated with disorders of deep placentation. *Am J Obstet Gynecol* 2011;204:193-201. doi:10.1016/j.ajog.2010.08.009

2 Than NG, Romero R, Tarca AL, et al. Integrated systems biology approach identifies novel maternal and placental pathways of preeclampsia. *Front Immunol* 2018;9:1661. doi:10.3389/fimmu.2018.01661

3 Turco MY, Gardner L, Hughes J, et al. Long-term, hormone-responsive organoid cultures of human endometrium in a chemically defined medium. *Nat Cell Biol* 2017;19:568-77. doi:10.1038/ncb3516

4 Haider S, Meinhardt G, Saleh L, et al. Self-renewing trophoblast organoids recapitulate the developmental program of the early human placenta. *Stem Cell Reports* 2018;11:537-51. doi:10.1016/j.stemcr.2018.07.004

5 Turco MY, Gardner L, Kay RG, et al. Trophoblast organoids as a model for maternal-fetal interactions during human placentation. *Nature* 2018;564:263-7. doi:10.1038/s41586-018-0753-3

6 Tranquilli AL, Brown MA, Zeeman GG, Dekker G, Sibai BM, Statements from the International Society for the Study of Hypertension in Pregnancy (ISSHP). The definition of severe and early-onset preeclampsia. *Pregnancy Hypertens* 2013;3:44-7. doi:10.1016/j.preghy.2012.11.001

7 Davey DA, MacGillivray I. The classification and definition of the hypertensive disorders of pregnancy. *Am J Obstet Gynecol* 1988;158:892-8. doi:10.1016/0002-9378(88)90090-7

8 Brown MA, Magee LA, Kenny LC, et al, International Society for the Study of Hypertension in Pregnancy (ISSHP). Hypertensive disorders of pregnancy: ISSHP classification, diagnosis, and management recommendations for international practice. *Hypertension* 2018;72:24-43. doi:10.1161/HYPERTENSIONAHA.117.10803

9 Staff AC, Benton SJ, von Dadelszen P, et al. Redefining preeclampsia using placenta-derived biomarkers. *Hypertension* 2013;61:932-42. doi:10.1161/HYPERTENSIONAHA.111.00250

10 Karumanchi SA. Angiogenic factors in preeclampsia: from diagnosis to therapy. *Hypertension* 2016;67:1072-9. doi:10.1161/HYPERTENSIONAHA.116.06421

11 Redman CW, Denson KW, Beilin LJ, Bolton FG, Stirrat GM. Factor-VIII consumption in pre-eclampsia. *Lancet* 1977;310:1249-52. doi:10.1016/S0140-6736(77)92661-7

12 Abalos E, Cuesta C, Grosso AL, Chou D, Say L. Global and regional estimates of preeclampsia and eclampsia: a systematic review.

13 Abalos E, Cuesta C, Carroli G, et al, WHO Multicountry Survey on Maternal and Newborn Health Research Network. Pre-eclampsia, eclampsia and adverse maternal and perinatal outcomes: a secondary analysis of the World Health Organization Multicountry Survey on Maternal and Newborn Health. *BJOG* 2014;121(Suppl 1):14-24. doi:10.1111/1471-0528.12629

14 Nakimuli A, Chazara O, Byamugisha J, et al. Pregnancy, parturition and preeclampsia in women of African ancestry. *Am J Obstet Gynecol* 2014;210:510-520.e1. doi:10.1016/j.ajog.2013.10.879

15 Jaatinen N, Ekholm E. Eclampsia in Finland; 2006 to 2010. *Acta Obstet Gynecol Scand* 2016;95:787-92. doi:10.1111/aogs.12882

16 Hodgins S, Tielsch J, Rankin K, Robinson A, Kearns A, Caglia J. A new look at care in pregnancy: simple, effective interventions for neglected populations. *PLoS One* 2016;11:e0160562. doi:10.1371/journal.pone.0160562

17 National Institute for Health and Care Excellence. Hypertension in pregnancy: the management of hypertensive disorders during pregnancy. 2010. https://www.nice.org.uk/guidance/cg107.

18 Duckitt K, Harrington D. Risk factors for pre-eclampsia at antenatal booking: systematic review of controlled studies. *BMJ* 2005;330:565. doi:10.1136/bmj.38380.674340.E0

19 Bartsch E, Medcalf KE, Park AL, Ray JG, High Risk of Pre-eclampsia Identification Group. Clinical risk factors for pre-eclampsia determined in early pregnancy: systematic review and meta-analysis of large cohort studies. *BMJ* 2016;353:i1753. doi:10.1136/bmj.i1753

20 Hernández-Díaz S, Toh S, Cnattingius S. Risk of pre-eclampsia in first and subsequent pregnancies: prospective cohort study. *BMJ* 2009;338:b2255. doi:10.1136/bmj.b2255

21 Schalekamp-Timmermans S, Arends LR, Alsaker E, et al, Global Pregnancy Collaboration. Fetal sex-specific differences in gestational age at delivery in pre-eclampsia: a meta-analysis. *Int J Epidemiol* 2017;46:632-42.

22 Gonzalez TL, Sun T, Koeppel AF, et al. Sex differences in the late first trimester human placenta transcriptome. *Biol Sex Differ* 2018;9:4. doi:10.1186/s13293-018-0165-y

23 Gong S, Johnson MD, Dopierala J, et al. Genome-wide oxidative bisulfite sequencing identifies sex-specific methylation differences in the human placenta. *Epigenetics* 2018;13:228-39. doi:10.1080/15592294.2018.1429857

24 Kalisch-Smith JI, Simmons DG, Dickinson H, Moritz KM. Review: Sexual dimorphism in the formation, function and adaptation of the placenta. *Placenta* 2017;54:10-6. doi:10.1016/j.placenta.2016.12.008

25 Eriksson JG, Kajantie E, Osmond C, Thornburg K, Barker DJ. Boys live dangerously in the womb. *Am J Hum Biol* 2010;22:330-5. doi:10.1002/ajhb.20995

26 Broere-Brown ZA, Schalekamp-Timmermans S, Hofman A, Jaddoe V, Steegers E. Fetal sex dependency of maternal vascular adaptation to pregnancy: a prospective population-based cohort study. *BJOG* 2016;123:1087-95. doi:10.1111/1471-0528.13519

27 Redman CW, Sargent IL, Staff AC. IFPA Senior Award Lecture: making sense of pre-eclampsia - two placental causes of preeclampsia? *Placenta* 2014;35(Suppl):S20-5. doi:10.1016/j.placenta.2013.12.008

28 Hubel CA. Oxidative stress in the pathogenesis of preeclampsia. *Proc Soc Exp Biol Med* 1999;222:222-35. doi:10.1046/j.1525-1373.1999.d01-139.x

29 Myatt L, Cui X. Oxidative stress in the placenta. *Histochem Cell Biol* 2004;122:369-82. doi:10.1007/s00418-004-0677-x

30 Tannetta D, Masliukaite I, Vatish M, Redman C, Sargent I. Update of syncytiotrophoblast derived extracellular vesicles in normal pregnancy and preeclampsia. *J Reprod Immunol* 2017;119:98-106. doi:10.1016/j.jri.2016.08.008

31 McLaughlin K, Zhang J, Lye SJ, Parker JD, Kingdom JC. Phenotypes of pregnant women who subsequently develop hypertension in pregnancy. *J Am Heart Assoc* 2018;7:e009595. doi:10.1161/JAHA.118.009595

32 Jauniaux E, Gulbis B, Burton GJ. The human first trimester gestational sac limits rather than facilitates oxygen transfer to the foetus-a review. *Placenta* 2003;24(Suppl A):S86-93. doi:10.1053/plac.2002.0932

33 Burton GJ, Watson AL, Hempstock J, Skepper JN, Jauniaux E. Uterine glands provide histiotrophic nutrition for the human fetus during the first trimester of pregnancy. *J Clin Endocrinol Metab* 2002;87:2954-9. doi:10.1210/jcem.87.6.8563

34 Spencer TE. Biological roles of uterine glands in pregnancy. *Semin Reprod Med* 2014;32:346-57. doi:10.1055/s-0034-1376354

35 Burton GJ. The John Hughes Memorial Lecture: Stimulation of early placental development through a trophoblast-endometrial

Eur J Obstet Gynecol Reprod Biol 2013;170:1-7. doi:10.1016/j.ejogrb.2013.05.005

dialogue. *J Equine Vet Sci* 2018;66:14-8. doi:10.1016/j.jevs.2018.03.003.

36 Burton GJ, Jauniaux E. The cytotrophoblastic shell and complications of pregnancy. *Placenta* 2017;60:134-9. doi:10.1016/j.placenta.2017.06.007

37 Conrad KP, Rabaglino MB, Post Uiterweer ED. Emerging role for dysregulated decidualization in the genesis of preeclampsia. *Placenta* 2017;60:119-29. doi:10.1016/j.placenta.2017.06.005

38 Garrido-Gomez T, Dominguez F, Quiñonero A, et al. Defective decidualization during and after severe preeclampsia reveals a possible maternal contribution to the etiology. *Proc Natl Acad Sci U S A* 2017;114:E8468-77. doi:10.1073/pnas.1706546114

39 Jauniaux E, Van Oppenraaij RH, Burton GJ. Obstetric outcome after early placental complications. *Curr Opin Obstet Gynecol* 2010;22:452-7. doi:10.1097/GCO.0b013e3283404e44

40 Okae H, Toh H, Sato T, et al. Derivation of human trophoblast stem cells. *Cell Stem Cell* 2018;22:50-63.e6. doi:10.1016/j.stem.2017.11.004

41 Pijnenborg R, Vercruysse L, Hanssens M. The uterine spiral arteries in human pregnancy: facts and controversies. *Placenta* 2006;27:939-58. doi:10.1016/j.placenta.2005.12.006

42 Whitley GS, Cartwright JE. Cellular and molecular regulation of spiral artery remodelling: lessons from the cardiovascular field. *Placenta* 2010;31:465-74. doi:10.1016/j.placenta.2010.03.002

43 Roberts VHJ, Morgan TK, Bednarek P, et al. Early first trimester uteroplacental flow and the progressive disintegration of spiral artery plugs: new insights from contrast-enhanced ultrasound and tissue histopathology. *Hum Reprod* 2017;32:2382-93. doi:10.1093/humrep/dex301

44 Pollheimer J, Vondra S, Baltayeva J, Beristain AG, Knöfler M. Regulation of Placental Extravillous Trophoblasts by the Maternal Uterine Environment. *Front Immunol* 2018;9:2597. doi:10.3389/fimmu.2018.02597

45 Vento-Tormo R, Efremova M, Botting RA, et al. Single-cell reconstruction of the early maternal-fetal interface in humans. *Nature* 2018;563:347-53. doi:10.1038/s41586-018-0698-6

46 Kadyrov M, Schmitz C, Black S, Kaufmann P, Huppertz B. Pre-eclampsia and maternal anaemia display reduced apoptosis and opposite invasive phenotypes of extravillous trophoblast. *Placenta* 2003;24:540-8. doi:10.1053/plac.2002.0946

47 Lyall F, Robson SC, Bulmer JN. Spiral artery remodeling and trophoblast invasion in preeclampsia and fetal growth restriction: relationship to clinical outcome. *Hypertension* 2013;62:1046-54. doi:10.1161/HYPERTENSIONAHA.113.01892

48 Burton GJ, Woods AW, Jauniaux E, Kingdom JC. Rheological and physiological consequences of conversion of the maternal spiral arteries for uteroplacental blood flow during human pregnancy. *Placenta* 2009;30:473-82. doi:10.1016/j.placenta.2009.02.009

49 Hutchinson ES, Brownbill P, Jones NW, et al. Utero-placental haemodynamics in the pathogenesis of pre-eclampsia. *Placenta* 2009;30:634-41. doi:10.1016/j.placenta.2009.04.011

50 Brosens I, Robertson WB, Dixon HG. The physiological response of the vessels of the placental bed to normal pregnancy. *J Pathol Bacteriol* 1967;93:569-79. doi:10.1002/path.1700930218

51 Robertson WB, Brosens I, Dixon HG. The pathological response of the vessels of the placental bed to hypertensive pregnancy. *J Pathol Bacteriol* 1967;93:581-92. doi:10.1002/path.1700930219

52 Aardema MW, Oosterhof H, Timmer A, van Rooy I, Aarnoudse JG. Uterine artery Doppler flow and uteroplacental vascular pathology in normal pregnancies and pregnancies complicated by pre-eclampsia and small for gestational age fetuses. *Placenta* 2001;22:405-11. doi:10.1053/plac.2001.0676

53 Khong TY, De Wolf F, Robertson WB, Brosens I. Inadequate maternal vascular response to placentation in pregnancies complicated by pre-eclampsia and by small-for-gestational age infants. *Br J Obstet Gynaecol* 1986;93:1049-59. doi:10.1111/j.1471-0528.1986.tb07830.x

54 Espinoza J, Romero R, Mee Kim Y, et al. Normal and abnormal transformation of the spiral arteries during pregnancy. *J Perinat Med* 2006;34:447-58. doi:10.1515/JPM.2006.089

55 Kim YM, Chaemsaithong P, Romero R, et al. The frequency of acute atherosis in normal pregnancy and preterm labor, preeclampsia, small-for-gestational age, fetal death and midtrimester spontaneous abortion. *J Matern Fetal Neonatal Med* 2015;28:2001-9. doi:10.3109/14767058.2014.976198

56 Sohlberg S, Mulic-Lutvica A, Lindgren P, Ortiz-Nieto F, Wikström AK, Wikström J. Placental perfusion in normal pregnancy and early and late preeclampsia: a magnetic resonance imaging study. *Placenta* 2014;35:202-6. doi:10.1016/j.placenta.2014.01.008

57 Everett TR, Lees CC. Beyond the placental bed: placental and systemic determinants of the uterine artery Doppler waveform. *Placenta* 2012;33:893-901. doi:10.1016/j.placenta.2012.07.011

58 Collins SL, Grant D, Black RS, Vellayan M, Impey L. Abdominal pregnancy: a perfusion confusion? *Placenta* 2011;32:793-5. doi:10.1016/j.placenta.2011.07.032

59 Rennie MY, Whiteley KJ, Adamson SL, Sled JG. Quantification of gestational changes in the uteroplacental vascular tree reveals vessel specific hemodynamic roles during pregnancy in mice. *Biol Reprod* 2016;95:43. doi:10.1095/biolreprod.116.140681

60 Clark AR, James JL, Stevenson GN, Collins SL. Understanding abnormal uterine artery Doppler waveforms: A novel computational model to explore potential causes within the utero-placental vasculature. *Placenta* 2018;66:74-81. doi:10.1016/j.placenta.2018.05.001

61 Sebire NJ. Implications of placental pathology for disease mechanisms; methods, issues and future approaches. *Placenta* 2017;52:122-6. doi:10.1016/j.placenta.2016.05.006

62 Falco ML, Sivanathan J, Laoreti A, Thilaganathan B, Khalil A. Placental histopathology associated with pre-eclampsia: systematic review and meta-analysis. *Ultrasound Obstet Gynecol* 2017;50:295-301. doi:10.1002/uog.17494

63 Pathak S, Sebire NJ, Hook L, et al. Relationship between placental morphology and histological findings in an unselected population near term. *Virchows Arch* 2011;459:11-20. doi:10.1007/s00428-011-1061-6

64 Sebire NJ, Goldin RD, Regan L. Term preeclampsia is associated with minimal histopathological placental features regardless of clinical severity. *J Obstet Gynaecol* 2005;25:117-8. doi:10.1080/014436105400041396

65 Ogge G, Chaiworapongsa T, Romero R, et al. Placental lesions associated with maternal underperfusion are more frequent in early-onset than in late-onset preeclampsia. *J Perinat Med* 2011;39:641-52. doi:10.1515/jpm.2011.098

66 Roberts JM, Escudero C. The placenta in preeclampsia. *Pregnancy Hypertens* 2012;2:72-83. doi:10.1016/j.preghy.2012.01.001

67 Nelson DB, Ziadie MS, McIntire DD, et al. Placental pathology suggesting that preeclampsia is more than one disease. *Am J Obstet Gynecol* 2014;210:66 e1-7.

68 Orabona R, Donzelli CM, Falchetti M, Santoro A, Valcamonico A, Frusca T. Placental histological patterns and uterine artery Doppler velocimetry in pregnancies complicated by early or late pre-eclampsia. *Ultrasound Obstet Gynecol* 2016;47:580-5. doi:10.1002/uog.15799

69 Alpoim PN, Godoi LC, Freitas LG, Gomes KB, Dusse LM. Assessment of L-arginine asymmetric 1 dimethyl (ADMA) in early-onset and late-onset (severe) preeclampsia. *Nitric Oxide* 2013;33:81-2. doi:10.1016/j.niox.2013.07.006

70 Kucur M, Tuten A, Oncul M, et al. Maternal serum apelin and YKL-40 levels in early and late-onset pre-eclampsia. *Hypertens Pregnancy* 2014;33:467-75. doi:10.3109/10641955.2014.944709

71 Levine RJ, Maynard SE, Qian C, et al. Circulating angiogenic factors and the risk of preeclampsia. *N Engl J Med* 2004;350:672-83. doi:10.1056/NEJMoa031884

72 Pinheiro CC, Rayol P, Gozzani L, et al. The relationship of angiogenic factors to maternal and neonatal manifestations of early-onset and late-onset preeclampsia. *Prenat Diagn* 2014;34:1084-92. doi:10.1002/pd.4432

73 Perales A, Delgado JL, de la Calle M, et al, STEPS investigators. sFlt-1/PlGF for prediction of early-onset pre-eclampsia: STEPS (Study of Early Pre-eclampsia in Spain). *Ultrasound Obstet Gynecol* 2017;50:373-82. doi:10.1002/uog.17373

74 Borges VTM, Zanati SG, Peraçoli MTS, et al. Maternal left ventricular hypertrophy and diastolic dysfunction and brain natriuretic peptide concentration in early- and late-onset pre-eclampsia. *Ultrasound Obstet Gynecol* 2018;51:519-23. doi:10.1002/uog.17495

75 He Y, Xu B, Song D, Yu F, Chen Q, Zhao M. Expression of the complement system's activation factors in plasma of patients with early/late-onset severe pre-eclampsia. *Am J Reprod Immunol* 2016;76:205-11. doi:10.1111/aji.12541

76 Tuten A, Oncul M, Kucur M, et al. Maternal serum copeptin concentrations in early- and late-onset pre-eclampsia. *Taiwan J Obstet Gynecol* 2015;54:350-4. doi:10.1016/j.tjog.2013.10.045

77 Sanhal CY, Can Kavcar M, Yucel A, Erkeneklı K, Erkaya S, Uygur D. Comparison of plasma fetuin A levels in patients with early-onset pre-eclampsia vs late-onset pre-eclampsia. *Eur J Obstet Gynecol Reprod Biol* 2016;200:108-12. doi:10.1016/j.ejogrb.2016.03.011

78 Dogan E, Demir SC, Gulec UK. Maternal soluble vascular cytoplasmic adhesion molecule-1 and fibronectin levels in early- and late-onset preeclamptic pregnancies. *Clin Exp Obstet Gynecol* 2014;41:681-4.

79 Chen Q, Wang Y, Zhao M, Hyett J, da Silva Costa F, Nie G. Serum levels of GDF15 are reduced in preeclampsia and the reduction is more profound in late-onset than early-onset cases. *Cytokine* 2016;83:226-30. doi:10.1016/j.cyto.2016.05.002

80 Teoh SS, Zhao M, Wang Y, Chen Q, Nie G. Serum HtrA1 is differentially regulated between early-onset and late-onset preeclampsia. *Placenta* 2015;36:990-5. doi:10.1016/j.placenta.2015.07.001

81 Ozel A, Davutoglu EA, Firat A, et al. Maternal serum irisin levels in early and late-onset pre-eclamptic and healthy pregnancies. *J Obstet Gynaecol* 2018;38:642-6. doi:10.1080/01443615.2017.1399260

82 Salimi S, Farajian-Mashhadi F, Naghavi A, et al. Different profile of serum leptin between early onset and late onset preeclampsia. *Dis Markers* 2014;2014:628476. doi:10.1155/2014/628476

83 Pillay P, Maharaj N, Moodley J, Mackraj I. Placental exosomes and pre-eclampsia: Maternal circulating levels in normal pregnancies and, early and late onset pre-eclamptic pregnancies. *Placenta* 2016;46:18-25. doi:10.1016/j.placenta.2016.08.078

84 Serdar Açıkgöz A, Tüten A, Öncül M, et al. Evaluation of maternal serum progranulin levels in normotensive pregnancies, and pregnancies with early- and late-onset preeclampsia. *J Matern Fetal Neonatal Med* 2016;29:2658-64.

85 Peraçoli JC, Bannwart-Castro CF, Romao M, et al. High levels of heat shock protein 70 are associated with pro-inflammatory cytokines and may differentiate early- from late-onset preeclampsia. *J Reprod Immunol* 2013;100:129-34. doi:10.1016/j.jri.2013.08.003

86 Ribeiro VR, Romao-Veiga M, Romagnoli GG, et al. Association between cytokine profile and transcription factors produced by T-cell subsets in early- and late-onset pre-eclampsia. *Immunology* 2017;152:163-73. doi:10.1111/imm.12757

87 Weber TM, Lackner HK, Roessler A, et al. Heart rate variability and baroreceptor reflex sensitivity in early- versus late-onset preeclampsia. *PLoS One* 2017;12:e0186521. doi:10.1371/journal.pone.0186521

88 Valensise H, Vasapollo B, Gagliardi G, Novelli GP. Early and late preeclampsia: two different maternal hemodynamic states in the latent phase of the disease. *Hypertension* 2008;52:873-80. doi:10.1161/HYPERTENSIONAHA.108.117358

89 Herzog EM, Eggink AJ, Reijnierse A, et al. Impact of early- and late-onset preeclampsia on features of placental and newborn vascular health. *Placenta* 2017;49:72-9. doi:10.1016/j.placenta.2016.11.014

90 Moldenhauer JS, Stanek J, Warshak C, Khoury J, Sibai B. The frequency and severity of placental findings in women with preeclampsia are gestational age dependent. *Am J Obstet Gynecol* 2003;189:1173-7. doi:10.1067/S0002-9378(03)00576-3

91 Egbor M, Ansari T, Morris N, Green CJ, Sibbons PD. Morphometric placental villous and vascular abnormalities in early- and late-onset pre-eclampsia with and without fetal growth restriction. *BJOG* 2006;113:580-9. doi:10.1111/j.1471-0528.2006.00882.x

92 Daglar K, Kirbas A, Timur H, Ozturk Inal Z, Danisman N. Placental levels of total oxidative and anti-oxidative status, ADAMTS-12 and decorin in early- and late-onset severe preeclampsia. *J Matern Fetal Neonatal Med* 2016;29:4059-64. doi:10.3109/14767058.2016.1154942

93 Herzog EM, Eggink AJ, Willemsen SP, et al. Early- and late-onset preeclampsia and the tissue-specific epigenome of the placenta and newborn. *Placenta* 2017;58:122-32. doi:10.1016/j.placenta.2017.08.070

94 Yung HW, Atkinson D, Campion-Smith T, Olovsson M, Charnock-Jones DS, Burton GJ. Differential activation of placental unfolded protein response pathways implies heterogeneity in causation of early- and late-onset pre-eclampsia. *J Pathol* 2014;234:262-76. doi:10.1002/path.4394

95 Fu J, Zhao L, Wang L, Zhu X. Expression of markers of endoplasmic reticulum stress-induced apoptosis in the placenta of women with early and late onset severe pre-eclampsia. *Taiwan J Obstet Gynecol* 2015;54:19-23. doi:10.1016/j.tjog.2014.11.002

96 Chen Y, Huang Y, Jiang R, Teng Y. Syncytiotrophoblast-derived microparticle shedding in early-onset and late-onset severe pre-eclampsia. *Int J Gynaecol Obstet* 2012;119:234-8. doi:10.1016/j.ijgo.2012.07.010

97 Vishnyakova PA, Volodina MA, Tarasova NV, et al. Mitochondrial role in adaptive response to stress conditions in preeclampsia. *Sci Rep* 2016;6:32410. doi:10.1038/srep32410

98 Liang M, Niu J, Zhang L, et al. Gene expression profiling reveals different molecular patterns in G-protein coupled receptor signaling pathways between early- and late-onset preeclampsia. *Placenta* 2016;40:52-9. doi:10.1016/j.placenta.2016.02.015

99 Zhang L, Yang H. Expression and localization of TLR4 and its negative regulator Tollip in the placenta of early-onset and late-onset preeclampsia. *Hypertens Pregnancy* 2012;31:218-27. doi:10.3109/10641955.2011.642434

100 Junus K, Centlow M, Wikström AK, Larsson I, Hansson SR, Olovsson M. Gene expression profiling of placentae from women with early- and late-onset pre-eclampsia: down-regulation of the angiogenesis-related genes ACVRL1 and EGFL7 in early-onset disease. *Mol Hum Reprod* 2012;18:146-55. doi:10.1093/molehr/gar067

101 Jones CJP, Fox H. An ultrastructural and ultrahistochemical study of the human placenta in maternal pre-eclampsia. *Placenta* 1980;1:61-76. doi:10.1016/S0143-4004(80)80016-6

102 Holland O, Dekker Nitert M, Gallo LA, Vejzovic M, Fisher JJ, Perkins AV. Review: Placental mitochondrial function and structure in gestational disorders. *Placenta* 2017;54:2-9. doi:10.1016/j.placenta.2016.12.012

103 Longtine MS, Chen B, Odibo AO, Zhong Y, Nelson DM. Villous trophoblast apoptosis is elevated and restricted to cytotrophoblasts in pregnancies complicated by preeclampsia, IUGR, or preeclampsia with IUGR. *Placenta* 2012;33:352-9. doi:10.1016/j.placenta.2012.01.017

104 Burton GJ, Jauniaux E. Pathophysiology of placental-derived fetal growth restriction. *Am J Obstet Gynecol* 2018;218(2S):S745-61. doi:10.1016/j.ajog.2017.11.577

105 Collett GP, Redman CW, Sargent IL, Vatish M. Endoplasmic reticulum stress stimulates the release of extracellular vesicles carrying danger-associated molecular pattern (DAMP) molecules. *Oncotarget* 2018;9:6707-17. doi:10.18632/oncotarget.24158

106 Tannetta D, Collett G, Vatish M, Redman C, Sargent I. Syncytiotrophoblast extracellular vesicles - Circulating biopsies reflecting placental health. *Placenta* 2017;52:134-8. doi:10.1016/j.placenta.2016.11.008

107 Roberts JM, Himes KP. Pre-eclampsia: Screening and aspirin therapy for prevention of pre-eclampsia. *Nat Rev Nephrol* 2017;13:602-4. doi:10.1038/nrneph.2017.121

108 Cindrova-Davies T, Yung HW, Johns J, et al. Oxidative stress, gene expression, and protein changes induced in the human placenta during labor. *Am J Pathol* 2007;171:1168-79. doi:10.2353/ajpath.2007.070528

109 Burton GJ, Sebire NJ, Myatt L, et al. Optimising sample collection for placental research. *Placenta* 2014;35:9-22. doi:10.1016/j.placenta.2013.11.005

110 Muralimanoharan S, Maloyan A, Myatt L. Evidence of sexual dimorphism in the placental function with severe preeclampsia. *Placenta* 2013;34:1183-9. doi:10.1016/j.placenta.2013.09.015

111 Veerbeek JH, Tissot Van Patot MC, Burton GJ, Yung HW. Endoplasmic reticulum stress is induced in the human placenta during labour. *Placenta* 2015;36:88-92. doi:10.1016/j.placenta.2014.11.005

112 Redman CW, Sargent IL. Latest advances in understanding preeclampsia. *Science* 2005;308:1592-4. doi:10.1126/science.1111726

113 Roberts JM, Lain KY. Recent Insights into the pathogenesis of pre-eclampsia. *Placenta* 2002;23:359-72. doi:10.1053/plac.2002.0819

114 Porto LB, Brandão AHF, Leite HV, Cabral ACV. Longitudinal evaluation of uterine perfusion, endothelial function and central blood flow in early onset pre-eclampsia. *Pregnancy Hypertens* 2017;10:161-4. doi:10.1016/j.preghy.2017.08.005

115 Roberts JM. Endothelial dysfunction in preeclampsia. *Semin Reprod Endocrinol* 1998;16:5-15. doi:10.1055/s-2007-1016248

116 Brosens I, Dixon HG, Robertson WB. Fetal growth retardation and the arteries of the placental bed. *Br J Obstet Gynaecol* 1977;84:656-63. doi:10.1111/j.1471-0528.1977.tb12676.x

117 Gerretsen G, Huisjes HJ, Elema JD. Morphological changes of the spiral arteries in the placental bed in relation to pre-eclampsia and fetal growth retardation. *Br J Obstet Gynaecol* 1981;88:876-81. doi:10.1111/j.1471-0528.1981.tb02222.x

118 Yung HW, Calabrese S, Hynx D, et al. Evidence of placental translation inhibition and endoplasmic reticulum stress in the etiology of human intrauterine growth restriction. *Am J Pathol* 2008;173:451-62. doi:10.2353/ajpath.2008.071193

119 Zhang K, Kaufman RJ. From endoplasmic-reticulum stress to the inflammatory response. *Nature* 2008;454:455-62. doi:10.1038/nature07203

120 Cindrova-Davies T, Fogarty NME, Jones CJP, Kingdom J, Burton GJ. Evidence of oxidative stress-induced senescence in mature, post-mature and pathological human placentas. *Placenta* 2018;68:15-22. doi:10.1016/j.placenta.2018.06.307

121 Johnson MR, Anim-Nyame N, Johnson P, Sooranna SR, Steer PJ. Does endothelial cell activation occur with intrauterine growth restriction?*BJOG* 2002;109:836-9. doi:10.1111/j.1471-0528.2002.01045.x

122 Sekizawa A, Jimbo M, Saito H, et al. Cell-free fetal DNA in the plasma of pregnant women with severe fetal growth restriction. *Am J Obstet Gynecol* 2003;188:480-4. doi:10.1067/mob.2003.27

123 Laivuori H, Gallaher MJ, Collura L, et al. Relationships between maternal plasma leptin, placental leptin mRNA and protein in normal pregnancy, pre-eclampsia and intrauterine growth restriction without pre-eclampsia. *Mol Hum Reprod* 2006;12:551-6. doi:10.1093/molehr/gal064

124 Goswami D, Tannetta DS, Magee LA, et al. Excess syncytiotrophoblast microparticle shedding is a feature of early-onset pre-eclampsia, but not normotensive intrauterine growth restriction. *Placenta* 2006;27:56-61. doi:10.1016/j.placenta.2004.11.007

125 Shibata E, Rajakumar A, Powers RW, et al. Soluble fms-like tyrosine kinase 1 is increased in preeclampsia but not in normotensive pregnancies with small-for-gestational-age neonates: relationship to circulating placental growth factor. *J Clin Endocrinol Metab* 2005;90:4895-903. doi:10.1210/jc.2004-1955

126 Chaiworapongsa T, Espinoza J, Gotsch F, et al. The maternal plasma soluble vascular endothelial growth factor receptor-1 concentration is elevated in SGA and the magnitude of the increase relates to Doppler abnormalities in the maternal and fetal circulation. *J Matern Fetal Neonatal Med* 2008;21:25-40. doi:10.1080/14767050701832833

127 Mizuuchi M, Cindrova-Davies T, Olovsson M, Charnock-Jones DS, Burton GJ, Yung HW. Placental endoplasmic reticulum stress negatively regulates transcription of placental growth factor via ATF4 and ATF6β: implications for the pathophysiology of human pregnancy complications. *J Pathol* 2016;238:550-61. doi:10.1002/path.4678

128 Cindrova-Davies T, Sanders DA, Burton GJ, Charnock-Jones DS. Soluble FLT1 sensitizes endothelial cells to inflammatory cytokines by antagonizing VEGF receptor-mediated signalling. *Cardiovasc Res* 2011;89:671-9. doi:10.1093/cvr/cvq346

129 Demirci O, Tuğrul AS, Dolgun N, Sözen H, Eren S. Serum lipids level assessed in early pregnancy and risk of pre-eclampsia. *J Obstet Gynaecol Res* 2011;37:1427-32. doi:10.1111/j.1447-0756.2011.01562.x

130 American College of Obstetricians and Gynecologists, Task Force on Hypertension in Pregnancy. Hypertension in pregnancy. Report of the American College of Obstetricians and Gynecologists' Task Force on Hypertension in Pregnancy. *Obstet Gynecol* 2013;122:1122-31.

131 Magee LA, Pels A, Helewa M, Rey E, von Dadelszen P, SOGC Hypertension Guideline Committee. Diagnosis, evaluation, and management of the hypertensive disorders of pregnancy: executive summary. *J Obstet Gynaecol Can* 2014;36:575-6. doi:10.1016/S1701-2163(15)30533-8

132 McCartney CP. Pathological anatomy of acute hypertension of pregnancy. *Circulation* 1964;30(suppl 2):2, 37-42. doi:10.1161/01.CIR.30.2S2.II-37

133 Piering WF, Garancis JG, Becker CG, Beres JA, Lemann JJr. Preeclampsia related to a functioning extrauterine placenta: report of a case and 25-year follow-up. *Am J Kidney Dis* 1993;21:310-3. doi:10.1016/S0272-6386(12)80751-7

134 Treloar SA, Cooper DW, Brennecke SP, Grehan MM, Martin NG. An Australian twin study of the genetic basis of preeclampsia and eclampsia. *Am J Obstet Gynecol* 2001;184:374-81. doi:10.1067/mob.2001.109400

135 Cnattingius S, Reilly M, Pawitan Y, Lichtenstein P. Maternal and fetal genetic factors account for most of familial aggregation of preeclampsia: a population-based Swedish cohort study. *Am J Med Genet A* 2004;130A:365-71. doi:10.1002/ajmg.a.30257

136 Skjaerven R, Vatten LJ, Wilcox AJ, Rønning T, Irgens LM, Lie RT. Recurrence of pre-eclampsia across generations: exploring fetal and maternal genetic components in a population based cohort. *BMJ* 2005;331:877. doi:10.1136/bmj.38555.462685.8F

137 McGinnis R, Steinthorsdottir V, Williams NO, et al, FINNPEC Consortium, GOPEC Consortium. Variants in the fetal genome near FLT1 are associated with risk of preeclampsia. *Nat Genet* 2017;49:1255-60. doi:10.1038/ng.3895

138 Medawar PB. *Some immunological and endocrinological problems raised by the evolution of viviparity in vertebrates. Society for experimental Biology.* New York Academic Press, 1953: 320-38.

139 Moffett A, Chazara O, Colucci F. Maternal allo-recognition of the fetus. *Fertil Steril* 2017;107:1269-72. doi:10.1016/j.fertnstert.2017.05.001

140 Hiby SE, Apps R, Sharkey AM, et al. Maternal activating KIRs protect against human reproductive failure mediated by fetal HLA-C2. *J Clin Invest* 2010;120:4102-10. doi:10.1172/JCI43998

141 Nakimuli A, Chazara O, Hiby SE, et al. A KIR B centromeric region present in Africans but not Europeans protects pregnant women from pre-eclampsia. *Proc Natl Acad Sci USA* 2015;112:845-50.

142 Moffett A, Colucci F. Co-evolution of NK receptors and HLA ligands in humans is driven by reproduction. *Immunol Rev* 2015;267:283-97. doi:10.1111/imr.12323

143 Lam MTC, Dierking E. Intensive Care Unit issues in eclampsia and HELLP syndrome. *Int J Crit Illn Inj Sci* 2017;7:136-41. doi:10.4103/IJCIIS.IJCIIS_33_17

144 Goldenberg RL, McClure EM, Macguire ER, Kamath BD, Jobe AH. Lessons for low-income regions following the reduction in hypertension-related maternal mortality in high-income countries. *Int J Gynaecol Obstet* 2011;113:91-5. doi:10.1016/j.ijgo.2011.01.002

145 Lisonkova S, Joseph KS. Incidence of preeclampsia: risk factors and outcomes associated with early- versus late-onset disease. *Am J Obstet Gynecol* 2013;209:544 e1-44.

146 Gudu W. Prodromal symptoms, health care seeking in response to symptoms and associated factors in eclamptic patients. *BMC Pregnancy Childbirth* 2017;17:87. doi:10.1186/s12884-017-1272-1

147 Velauthar L, Plana MN, Kalidindi M, et al. First-trimester uterine artery Doppler and adverse pregnancy outcome: a meta-analysis involving 55,974 women. *Ultrasound Obstet Gynecol* 2014;43:500-7. doi:10.1002/uog.13275

148 O'Gorman N, Wright D, Poon LC, et al. Accuracy of competing-risks model in screening for pre-eclampsia by maternal factors and biomarkers at 11-13 weeks' gestation. *Ultrasound Obstet Gynecol* 2017;49:751-5. doi:10.1002/uog.17399

149 Chappell LC, Duckworth S, Seed PT, et al. Diagnostic accuracy of placental growth factor in women with suspected preeclampsia: a prospective multicenter study. *Circulation* 2013;128:2121-31. doi:10.1161/CIRCULATIONAHA.113.003215

150 Zeisler H, Llurba E, Chantraine F, et al. Predictive value of the sFlt-1:PlGF ratio in women with suspected preeclampsia. *N Engl J Med* 2016;374:13-22. doi:10.1056/NEJMoa1414838

151 Rolnik DL, Wright D, Poon LC, et al. Aspirin versus placebo in pregnancies at high risk for preterm preeclampsia. *N Engl J Med* 2017;377:613-22. doi:10.1056/NEJMoa1704559

152 Roberts JM, Balk JL, Bodnar LM, Belizán JM, Bergel E, Martinez A. Nutrient involvement in preeclampsia. *J Nutr* 2003;133(Suppl 2):1684S-92S. doi:10.1093/jn/133.5.1684S

153 Thangaratinam S, Rogozinska E, Jolly K, et al. Effects of interventions in pregnancy on maternal weight and obstetric outcomes: meta-analysis of randomised evidence. *BMJ* 2012;344:e2088. doi:10.1136/bmj.e2088

154 Klonoff-Cohen HS, Cross JL, Pieper CF. Job stress and preeclampsia. *Epidemiology* 1996;7:245-9. doi:10.1097/00001648-199605000-00005

155 Duley L, Henderson-Smart DJ, Meher S, King JF. Antiplatelet agents for preventing pre-eclampsia and its complications. *Cochrane Database Syst Rev* 2007;2:CD004659. doi:10.1002/14651858.CD004659.pub2

156 Bibbins-Domingo K, Grossman DC, Curry SJ, et al, US Preventive Services Task Force. Screening for Preeclampsia: US Preventive Services Task Force Recommendation Statement. *JAMA* 2017;317:1661-7. doi:10.1001/jama.2017.3439

157 Hofmeyr GJ, Lawrie TA, Atallah AN, Torloni MR. Calcium supplementation during pregnancy for preventing hypertensive disorders and related problems. *Cochrane Database Syst Rev* 2018;10:CD001059. doi:10.1002/14651858.CD001059.pub5

158 Rumbold A, Duley L, Crowther CA, Haslam RR. Antioxidants for preventing pre-eclampsia. *Cochrane Database Syst Rev* 2008;1:CD004227.

159 Poston L, Chappell L, Seed P, Shennan A. Biomarkers of oxidative stress in pre-eclampsia. *Pregnancy Hypertens* 2011;1:22-7. doi:10.1016/j.preghy.2010.10.009

160 Wolf G. How an increased intake of alpha-tocopherol can suppress the bioavailability of gamma-tocopherol. *Nutr Rev* 2006;64:295-9. doi:10.1111/j.1753-4887.2006.tb00213.x

161 Cindrova-Davies T. The therapeutic potential of antioxidants, ER chaperones, NO and H2S donors, and statins for treatment of preeclampsia. *Front Pharmacol* 2014;5:119. doi:10.3389/fphar.2014.00119

162 Catov JM, Nohr EA, Bodnar LM, Knudson VK, Olsen SF, Olsen J. Association of periconceptional multivitamin use with reduced risk of preeclampsia among normal-weight women in the Danish National Birth Cohort. *Am J Epidemiol* 2009;169:1304-11. doi:10.1093/aje/kwp052

163 Bodnar LM, Tang G, Ness RB, Harger G, Roberts JM. Periconceptional multivitamin use reduces the risk of preeclampsia. *Am J Epidemiol* 2006;164:470-7. doi:10.1093/aje/kwj218

164 Roberts JM, Bell MJ. If we know so much about preeclampsia, why haven't we cured the disease? *J Reprod Immunol* 2013;99:1-9. doi:10.1016/j.jri.2013.05.003

165 Brown MA, Magee LA, Kenny LC, et al. International Society for the Study of Hypertension in Pregnancy (ISSHP). The hypertensive disorders of pregnancy: ISSHP classification, diagnosis & management recommendations for international practice. *Pregnancy Hypertens* 2018;13:291-310. doi:10.1016/j.preghy.2018.05.004

166 Lucas MJ, Leveno KJ, Cunningham FG. A comparison of magnesium sulfate with phenytoin for the prevention of eclampsia. *N Engl J Med* 1995;333:201-5. doi:10.1056/NEJM199507273330401

167 Duley L, Eclampsia Trial Collaborative Group. Magnesium sulphate in eclampsia. *Lancet* 1998;352:67-8. doi:10.1016/S0140-6736(05)79550-7

168 Duley L, Henderson-Smart DJ, Walker GJ, Chou D. Magnesium sulphate versus diazepam for eclampsia. *Cochrane Database Syst Rev* 2010;12:CD000127.

169 Duley L, Gülmezoglu AM. Magnesium sulfate compared with lytic cocktail for women with eclampsia. *Int J Gynaecol Obstet* 2002;76:3-8. doi:10.1016/S0020-7292(01)00559-8

170 Bellamy L, Casas JP, Hingorani AD, Williams DJ. Pre-eclampsia and risk of cardiovascular disease and cancer in later life: systematic review and meta-analysis. *BMJ* 2007;335:974. doi:10.1136/bmj.39335.385301.BE

171 Mongraw-Chaffin ML, Cirillo PM, Cohn BA. Preeclampsia and cardiovascular disease death: prospective evidence from the child health and development studies cohort. *Hypertension* 2010;56:166-71. doi:10.1161/HYPERTENSIONAHA.110.150078

172 den Ruijter H, Pasterkamp G, Rutten FH, et al. Heart failure with preserved ejection fraction in women: the Dutch Queen of Hearts program. *Neth Heart J* 2015;23:89-93. doi:10.1007/s12471-014-0613-1

173 Melchiorre K, Sutherland GR, Liberati M, Thilaganathan B. Preeclampsia is associated with persistent postpartum cardiovascular impairment. *Hypertension* 2011;58:709-15. doi:10.1161/HYPERTENSIONAHA.111.176537

174 Hasan KM, Manyonda IT, Ng FS, Singer DR, Antonios TF. Skin capillary density changes in normal pregnancy and pre-eclampsia. *J Hypertens* 2002;20:2439-43. doi:10.1097/00004872-200212000-00024

175 Romundstad PR, Magnussen EB, Smith GD, Vatten LJ. Hypertension in pregnancy and later cardiovascular risk: common antecedents?*Circulation* 2010;122:579-84. doi:10.1161/CIRCULATIONAHA.110.943407

176 Gentry MB, Dias JK, Luis A, Patel R, Thornton J, Reed GL. African-American women have a higher risk for developing peripartum cardiomyopathy. *J Am Coll Cardiol* 2010;55:654-9. doi:10.1016/j.jacc.2009.09.043

177 Ersbøll AS, Johansen M, Damm P, Rasmussen S, Vejlstrup NG, Gustafsson F. Peripartum cardiomyopathy in Denmark: a retrospective, population-based study of incidence, management and outcome. *Eur J Heart Fail* 2017;19:1712-20. doi:10.1002/ejhf.882

178 Mosca L, Benjamin EJ, Berra K, et al, American Heart Association. Effectiveness-based guidelines for the prevention of cardiovascular disease in women--2011 update: a guideline from the American Heart Association. *J Am Coll Cardiol* 2011;57:1404-23. doi:10.1016/j.jacc.2011.02.005

179 Staff AC, Redman CW, Williams D, et al, Global Pregnancy Collaboration (CoLab). Pregnancy and long-term maternal cardiovascular health: progress through harmonization of research cohorts and biobanks. *Hypertension* 2016;67:251-60. doi:10.1161/HYPERTENSIONAHA.115.06357

180 Roberts JM, Catov JM. Pregnancy is a screening test for later life cardiovascular disease: now what? Research recommendations. *Womens Health Issues* 2012;22:e123-8. doi:10.1016/j.whi.2012.01.001

181 Baecke M, Spaanderman ME, van der Werf SP. Cognitive function after pre-eclampsia: an explorative study. *J Psychosom Obstet Gynaecol* 2009;30:58-64. doi:10.1080/01674820802546212

182 Postma IR, Bouma A, de Groot JC, Aukes AM, Aarnoudse JG, Zeeman GG. Cerebral white matter lesions, subjective cognitive failures, and objective neurocognitive functioning: A follow-up study in women after hypertensive disorders of pregnancy. *J Clin Exp Neuropsychol* 2016;38:585-98. doi:10.1080/13803395.2016.1143453

183 Troisi R, Innes KE, Roberts JM, Hoover RN. Preeclampsia and maternal breast cancer risk by offspring gender: do elevated androgen concentrations play a role?*Br J Cancer* 2007;97:688-90. doi:10.1038/sj.bjc.6603921

184 Davis EF, Lazdam M, Lewandowski AJ, et al. Cardiovascular risk factors in children and young adults born to preeclamptic pregnancies: a systematic review. *Pediatrics* 2012;129:e1552-61. doi:10.1542/peds.2011-3093

185 Kajantie E, Eriksson JG, Osmond C, Thornburg K, Barker DJ. Pre-eclampsia is associated with increased risk of stroke in the adult offspring: the Helsinki birth cohort study. *Stroke* 2009;40:1176-80. doi:10.1161/STROKEAHA.108.538025

186 Tuovinen S, Räikkönen K, Pesonen AK, et al. Hypertensive disorders in pregnancy and risk of severe mental disorders in the offspring in adulthood: the Helsinki Birth Cohort Study. *J Psychiatr Res* 2012;46:303-10. doi:10.1016/j.jpsychires.2011.11.015

187 Burton GJ, Fowden AL, Thornburg KL. Placental origins of chronic disease. *Physiol Rev* 2016;96:1509-65. doi:10.1152/physrev.00029.2015

188 Alsnes IV, Vatten LJ, Fraser A, et al. Hypertension in pregnancy and offspring cardiovascular risk in young adulthood: prospective and sibling studies in the HUNT study (Nord-Trondelag Health Study) in Norway. *Hypertension* 2017;69:591-8. doi:10.1161/HYPERTENSIONAHA.116.08414

189 Costantine MM, Cleary K, Hebert MF, et al. Safety and pharmacokinetics of pravastatin used for the prevention of preeclampsia in high-risk pregnant women: a pilot randomized controlled trial.*Am J Obstet Gynecol* 2016;214:e1-17.

190 Sharp A, Cornforth C, Jackson R, et al, STRIDER group. Maternal sildenafil for severe fetal growth restriction (STRIDER): a multicentre, randomised, placebo-controlled, double-blind trial. *Lancet Child Adolesc Health* 2018;2:93-102. doi:10.1016/S2352-4642(17)30173-6

191 Groom KM, Ganzevoort W, Alfirevic Z, Lim K, Papageorghiou AT, STRIDER Consortium. Clinicians should stop prescribing sildenafil for fetal growth restriction (FGR): comment from the STRIDER Consortium. *Ultrasound Obstet Gynecol* 2018;52:295-6. doi:10.1002/uog.19186

192 Dorniak-Wall T, Grivell RM, Dekker GA, Hague W, Dodd JM. The role of L-arginine in the prevention and treatment of pre-eclampsia: a systematic review of randomised trials. *J Hum Hypertens* 2014;28:230-5. doi:10.1038/jhh.2013.100

193 Gui S, Jia J, Niu X, et al. Arginine supplementation for improving maternal and neonatal outcomes in hypertensive disorder of pregnancy: a systematic review. *J Renin Angiotensin Aldosterone Syst* 2014;15:88-96. doi:10.1177/1470320313475910

194 Thadhani R, Hagmann H, Schaarschmidt W, et al. Removal of soluble Fms-like tyrosine kinase-1 by dextran sulfate apheresis in preeclampsia. *J Am Soc Nephrol* 2016;27:903-13. doi:10.1681/ASN.2015020157

195 Turanov AA, Lo A, Hassler MR, et al. RNAi modulation of placental sFLT1 for the treatment of preeclampsia. *Nat Biotechnol* 2018;36:1164-73. doi:10.1038/nbt.4297

196 Gunnarsson R, Åkerström B, Hansson SR, Gram M. Recombinant alpha-1-microglobulin: a potential treatment for preeclampsia. *Drug Discov Today* 2017;22:736-43. doi:10.1016/j.drudis.2016.12.005

197 Powers RW, Roberts JM, Plymire DA, et al. Low placental growth factor across pregnancy identifies a subset of women with preterm preeclampsia: type 1 versus type 2 preeclampsia?*Hypertension* 2012;60:239-46. doi:10.1161/HYPERTENSIONAHA.112.191213

198 Myatt L, Roberts JM, Redman CWG, Global Pregnancy Collaboration (CoLab). Availability of COLLECT, a database for pregnancy and placental research studies worldwide. *Placenta* 2017;57:223-4. doi:10.1016/j.placenta.2017.07.014

199 Ghosh G, Grewal J, Männistö T, et al. Racial/ethnic differences in pregnancy-related hypertensive disease in nulliparous women. *Ethn Dis* 2014;24:283-9.

200 Maric-Bilkan C, Abrahams VM, Arteaga SS, et al. Research recommendations from the National Institutes of Health Workshop on predicting, preventing, and treating preeclampsia. *Hypertension* 2019;73:757-66. doi:10.1161/HYPERTENSIONAHA.118.11644

201 Goldenberg RL, McClure EM, Macguire ER, Kamath BD, Jobe AH. Lessons for low-income regions following the reduction in hypertension-related maternal mortality in high-income countries. *Int J Gynaecol Obstet* 2011;113:91-5.

202 Abalos E, Cuesta C, Carroli G, et al. WHO multicountry survey on maternal and newborn health research network. Pre-eclampsia, eclampsia and adverse maternal and perinatal outcomes: a secondary analysis of the World Health Organization multicountry survey on maternal and newborn health. *BJOG* 2014;121(Suppl 1):14-24.

203 Knight M, Nair M, Tuffnell D, Shakespeare J, Kenyon S, Kurinczuk JJ (eds.). on behalf of MBRRACE-UK. Saving lives, improving mothers' care—lessons learned to inform maternity care from the UK and Ireland confidential enquiries into maternal deaths and morbidity 2013−15. Oxford: National PerinatalEpidemiology Unit, University of Oxford 2017. https://www.npeu.ox.ac.uk/downloads/files/mbrrace-uk/reports/MBRRACE-UK%20Maternal%20Report%202017%20-%20Web.pdf