Review

Yolanda Correia*, Julia Scheel*, Shailendra Gupta and Keqing Wang

# Placental mitochondrial function as a driver of angiogenesis and placental dysfunction

https://doi.org/10.1515/hsz-2021-0121
Received January 22, 2021; accepted June 9, 2021;
published online July 5, 2021

**Abstract:** The placenta is a highly vascularized and complex foetal organ that performs various tasks, crucial to a healthy pregnancy. Its dysfunction leads to complications such as stillbirth, preeclampsia, and intrauterine growth restriction. The specific cause of placental dysfunction remains unknown. Recently, the role of mitochondrial function and mitochondrial adaptations in the context of angiogenesis and placental dysfunction is getting more attention. The required energy for placental remodelling, nutrient transport, hormone synthesis, and the reactive oxygen species leads to oxidative stress, stemming from mitochondria. Mitochondria adapt to environmental changes and have been shown to adjust their oxygen and nutrient use to best support placental angiogenesis and foetal development. Angiogenesis is the process by which blood vessels form and is essential for the delivery of nutrients to the body. This process is regulated by different factors, pro-angiogenic factors and anti-angiogenic factors, such as sFlt-1. Increased circulating sFlt-1 levels have been linked to different preeclamptic phenotypes. One of many effects of increased sFlt-1 levels, is the dysregulation of mitochondrial function. This review covers mitochondrial adaptations during placentation, the importance of the anti-angiogenic factor sFlt-1in placental dysfunction and its role in the dysregulation of mitochondrial function.

**Keywords:** computational modelling; endothelial dysfunction; mitochondrial dysfunction; preeclampsia; systems biology; vascular deregulation.

## Introduction: preeclampsia

Preeclampsia (PE) is a multisystemic disorder (Lambert et al. 2014) that affects 1–8% of pregnancies (Duley 2009). Annually, 76,000 maternal deaths and 500,000 perinatal deaths worldwide can be attributed to PE (Khan et al. 2006) and it is accompanied by severe long-term burdens. PE is defined as *de novo* onset of hypertension (≥160/110 mmHg) (Brown et al. 2018) and proteinuria (spot urine protein/creatinine > 30 mg/mmol or >300 mg/day) (Tranquilli et al. 2014; Trogstad et al. 2011), after 20 weeks of gestation. The exact pathophysiology and phenotype subtypes of PE are poorly understood.

Although the placenta is understood to be the main actor in PE, which should thus be "cured" after delivery of the placenta, recent studies showed that preeclamptic women have an increased chance of developing various long-term cardiovascular diseases. PE strongly correlates with subsequent development of hypertension, ischaemic heart disease, stroke and venous thromboembolism (Bellamy et al. 2007). Correspondingly, several predisposing factors, such as multifetal pregnancies, chronic high blood pressure, personal or family history of PE, pre-existing diabetes, thrombophilia and obesity, are associated with increased risks of developing PE (English et al. 2015).

PE is a multifactorial disorder and the exact aetiology is still unknown. The disruption of endothelial homeostasis, abnormal placentation, excessive inflammation, and imbalance of angiogenic factors are key features of PE (Apicella et al. 2019; Possomato-Vieira and Khalil 2016; Sánchez-Aranguren et al. 2014).

To date, there are no effective treatments to prevent PE, partly due to the lack of clear knowledge behind the pathophysiology of PE. The current therapeutic strategies for PE mainly focus on alleviating the symptoms in order to delay the necessity for delivery. However, this strategy

**Yolanda Correia and Julia Scheel** contributed equally to this article.

***Corresponding authors: Yolanda Correia** Aston Medical School, College of Health & Life Sciences, Aston University, Aston Triangle, Birmingham B4 7ET, UK, E-mail: y.correia@aston.ac.uk; and **Julia Scheel**, Department of Systems Biology and Bioinformatics, University of Rostock, D-18051 Rostock, Germany, E-mail: julia.scheel@uni-rostock.de. https://orcid.org/0000-0002-0034-7755
**Shailendra Gupta,** Department of Systems Biology and Bioinformatics, University of Rostock, D-18051 Rostock, Germany
**Keqing Wang,** Aston Medical School, College of Health & Life Sciences, Aston University, Aston Triangle, Birmingham B4 7ET, UK

often leads to preterm delivery in most cases (Perry et al. 2018). Therefore, there is an urgent need for a better understanding and better management of PE. Various treatments involving statins (predominantly pravastatin), aspirin, calcium and metformin have been explored to prevent the onset of PE and to treat its associated symptoms. However, despite the extensive research into the potential causes and mechanisms of PE, there is no cure for PE.

# Pathogenesis of preeclampsia

## Two-stage theory and preeclampsia

It is widely accepted that PE is a placental disease occurring in two stages: during stage 1, synonymous of abnormal placentation happens early in the first trimester leading to decreased placental perfusion, and subsequent stage 2, a "maternal syndrome in the later second and third trimesters characterized by an excess of antiangiogenic factors" (Lain and Roberts 2002; Roberts and Escudero 2012; Roberts and Gammill 2005; Roberts and Hubel 2009; Roberts et al. 1989; Staff 2019).

The maternal uterus undergoes extreme structural changes during pregnancy. The molecular processes of placentation are meticulously coordinated. Any disturbance of the equilibrium within the fetomaternal interface, such as increased inflammatory responses can have detrimental effects (Mayrink et al. 2018).

Maternal spiral arteries undergo intensive remodelling in pregnancy resulting in increased blood flow to intervillous space to ensure efficient supply to the foetus (Sweeney and Foldes 2018). During the first nine weeks of pregnancy, extravillous trophoblasts (EVT) invade spiral arteries and form plugs, partially blocking the flow of oxygenated blood to the placental surface during early gestation. This creates a hypoxic environment, which is essential for accurate trophoblast differentiation and placentation, respectively. During the first trimester, these trophoblast plugs progressively loosen, until their structure is disintegrated (Figure 1). The blood flow then significantly increases around week 13 of gestation (Moser et al. 2015). Inadequate spiral artery remodelling can lead to dysregulated utero-placental circulation, increased oxidative stress, as well as a release of placental factors into the maternal circulation (Redman and Sargent 2010). As inadequate spiral artery remodelling also occurs in other diseases such as foetal growth restriction and foetal death and does not cause PE on its own, it is considered one of many factors adding to the phenotype of PE, rather than its main inducement (Burke et al. 2010).

Stem cell trophoblasts differentiate into cytotrophoblasts (CT), which first invade the decidua compacta and then the gland containing decidua spongiosa. The remodelling and additional recruitment of natural killer cells (NK), dendritic cells (DC) and macrophages (M) from the myometrium lead to the thickening of the decidua. Cytotrophoblasts differentiate into extravillous trophoblasts (EVT) and villous trophoblasts (VT). Decidual EC are replaced by endovascular trophoblasts (a subgroup of EVTs), which break through the walls of the uterine spiral arteries, while smooth muscle cells are replaced by connective tissue and fibroblasts. This process ensures the widening of spiral arteries during pregnancy and a low resistance to blood flow, hence sufficient supplementation of villi (James et al. 2017; Vento-tormo et al. 2018).

## Endothelial dysfunction in preeclampsia

The endothelium is crucial in controlling vascular tone and permeability through autocrine, paracrine and endocrine signalling (Sandoo et al. 2015). The proliferation of endothelial cells (EC) depends on mitochondrial biogenesis and dynamics, which are influenced by a variety of factors such as oxygen, haemodynamic changes, and nutrients. EC can become activated in response to different stimuli, most commonly inflammatory cytokines such as TNFα and IL-6 (Bailey et al. 2017; Chen et al. 2009; Laresgoiti-Servitje 2013; Liao 2013; Roberts 1989; Szarka et al. 2010; Videm and Albrigtsen 2008). Widespread endothelial dysfunction (ED) is a hallmark of PE (Maynard et al. 2003; Roberts 1989; Sánchez-Aranguren 2014; Wang and Walsh 1998) and women at high risk of developing PE during pregnancy demonstrate ED before the onset of clinical symptoms (Muller et al. 1993). Increased levels of endothelial activation markers have been reported in women who develop PE. Plasma levels of adhesion cell molecules are significantly increased in women with PE compared to normotensive pregnancy (Carty 2012). Soluble E-selectin was significantly higher in women at 12–16 weeks' gestation who went on to develop PE compared to women who had a healthy pregnancy (Carty 2012). Significant elevations in E-selectin are also found to be higher in the postpartum period in women with PE (Papakonstantinou et al. 2011). Furthermore, endothelial activation and dysfunction, as well as the appearance of cardiovascular diseases, have been detected after a pregnancy complicated with PE up to 20–25 years post-partum (Evans et al. 2011; Wong et al. 2003).

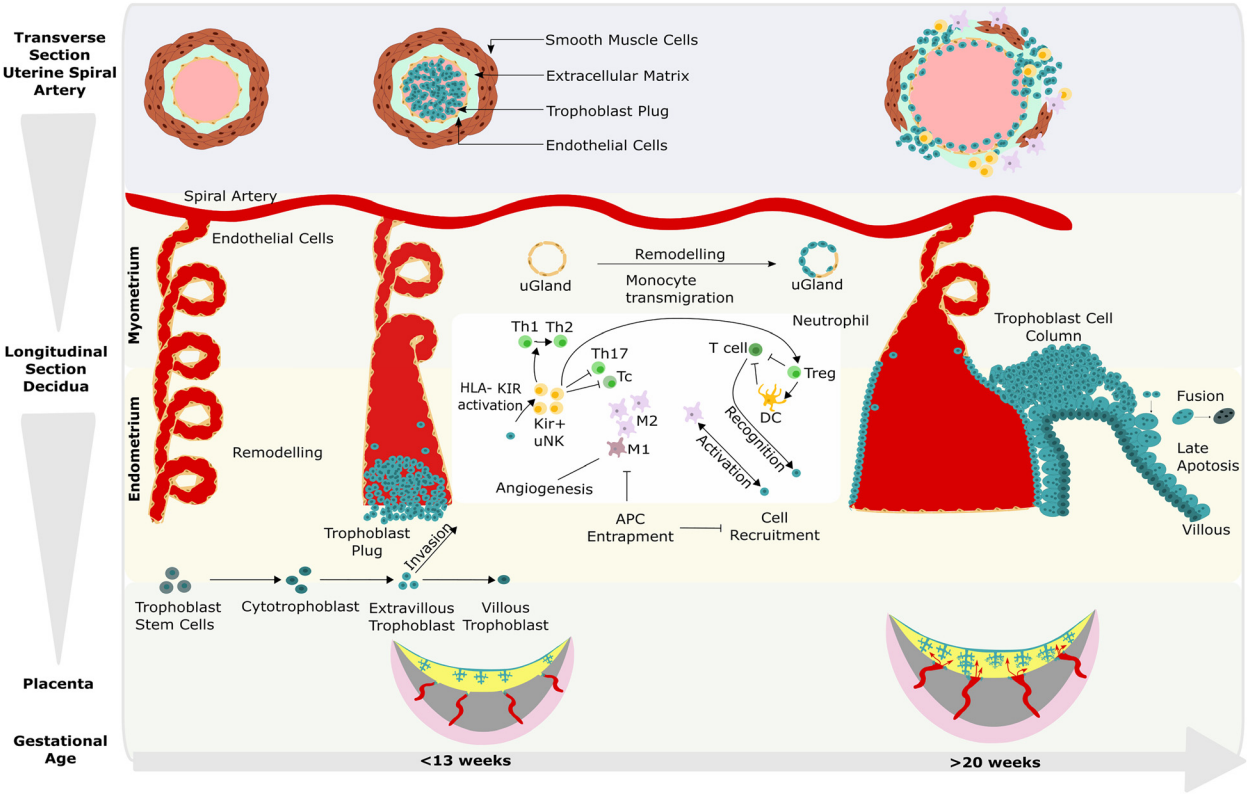



**Figure 1:** Spiral artery remodelling and biological processes in placenta development according to gestational age.
Placental development processes are meticulously coordinated. Any disturbance of the environment through oxidative stress or mitochondrial deregulation can lead to subsequent deregulation of these processes and placental dysfunction. Trophoblast stem cells differentiate into cytotrophoblasts and then extravillous trophoblasts, which invade the endometrium. Extravillous trophoblasts form plugs creating a hypoxic environment ideal for spiral artery remodelling. After 13 weeks gestational age, the plug progressively loosens until the blood flow is uninhibited and gas and nutrient exchange across the fetomaternal interface.

Vascular endothelial growth factor (VEGF) is critical in the process of new blood vessel generation and in the endothelial homeostasis. One of the mechanisms of ED involves the release of the sFlt-1 soluble fms-like tyrosine kinase (sFlt-1 or sVEGFR1), which is a circulating anti-angiogenic protein and an endogenous inhibitor of VEGF (Samie Omran and Mohammed Osman 2016). Circulating levels of antiangiogenic factors released by the placenta contribute to maternal ED and to the clinical onset of PE. Overexpression of sFlt-1 induces ED in mice where PE symptoms are present (Bergmann et al. 2010). Circulating levels of sFlt-1 and sEng are significantly increased in PE and elevation of these factors correlates with disease severity (Hertig et al. 2004; Levine et al. 2005). Furthermore, it's demonstrated that hypoxic placentas secrete increased levels of VEGF. However, secretion of sFlt-1 increases to a greater extent than VEGF secretion, generating an imbalance of anti-angiogenic factors under the hypoxic condition (Chaiworapongsa et al. 2004; Forsythe et al. 1996; Hertig 2004; Koga et al. 2003; Levine 2005; Maynard 2003).

Using the supplementation of VEGF or PlGF as the buffering mechanisms for excess sFlt-1 has been tested in different animal PE models (Bergmann 2010; Chen et al. 2008; Logue et al. 2017; Miquerol et al. 2000; Rees et al. 1990; Woods et al. 2011). For example, adenoviral delivery of VEGF in pregnant BPH/5 mice inhibited the spontaneous development of PE-like symptoms in BPH/5 Mice (Williams et al. 2013; Woods 2011). Adenoviral delivery of VEGF (Gilbert et al. 2007) in sFlt-1 overexpressing pregnant mice led to reduced blood pressure and kidney injury (Bergmann 2010). Additionally, different forms of PlGF have also been tested in different animal models (George et al. 2015; Makris et al. 2007; Spradley et al. 2016; Suzuki et al. 2009; Watanabe and Dvorak 1997). It has been demonstrated that the administration of placental growth factor-2 (PIGF-2) and VEGF in pregnant mice overexpressing sFlt-1, decreased mean blood pressure (Suzuki 2009). Spradley et al. looked at the five-day infusion of recombinant human form of PIGF at gestational day 14 on pregnant rats that had undergone Reduced Uterine Perfusion Pressure (RUPP)

procedure and demonstrated an abolishment of the RUPP after effects, such as an increase in glomerular filtration rate, a decrease in the mean arterial blood pressure and a decrease of plasma sFlt-1 (Spradley 2016). Furthermore, removal of circulating plasma sFlt-1 by apheresis with a plasma-specific dextran sulphate showed a significant reduction in proteinuria, improved blood pressure and prolonged the pregnancy (Thadhani et al. 2011, 2016). These studies further highlight angiogenic imbalance is the major 'culprit' of PE.

## Genetic aspects of preeclampsia

In the same way that ED has been associated with PE, vasoactive proteins, thrombophilia and hypofibrinolysis, oxidative stress, lipid metabolism and immunogenetics have also been linked to PE ("The Genetics of Pre-Eclampsia and Other Hypertensive Disorders of Pregnancy" 2011). In a 2017 study (Vishnyakova et al. 2017) the myometrium of women with a normotensive pregnancy and women with a preeclamptic pregnancy were compared and several significant differences were observed. These significant differences presented in the form of an increase in both the levels of antioxidant enzyme markers of mitochondrial biogenesis (VDAC1, TFAM, hexokinase 1, PGC-1α and PGC-1β) and autophagy proteins (LC3A) in the preeclamptic myometrium when compared to normotensive myometrium (Vishnyakova 2017). This study, alongside many others (Navarro-Yepes et al. 2014; "The Genetics of Pre-Eclampsia and Other Hypertensive Disorders of Pregnancy" 2011; Wang 1998; Zorov et al. 2014) provides evidence that molecular/genetic changes occur in the maternal myometrium with the development of PE. As shown here in the case of PE, complex diseases can have various interacting causes. In the age of personalized medicine, the genetic basis of diseases becomes more and more important. Some genes may predispose someone to develop a disease, whereas rare genetic variants may contribute more directly to a disease (Carolina Sanchez-Aranguren et al. 2020). Therefore, to date no specific gene has been universally identified as a susceptibility gene for PE. However, according to DisGeNet, a dedicated database repository curated by experts for diseases and their associated genes based on experiments, scientific literature, 166 genes and 14 gene variants have been associated with PE (Supplementary Material 1). *ZNF295-AS1, ASB4, FLT1P1, KIR3DL3, GCM1* are the genes with the highest disease specificity index score for PE and are candidates for transgenic PE animal models (Table 1).

Impaired vascularization and ED have been shown in PE. Although these phenotypes often coincide, only a small group of genes identified in their respective gene-disease associations overlap. Cardiovascular complications are a common long-term complication of preeclamptic pregnancies. Although pregnancy associated cardiovascular complications and predisposed maternal conditions such as hypertension are considered separate diseases, it is useful to compare overlapping disease-gene associations as they are risk factors for PE. Given the sensitive topic and vulnerability of affected groups, the number of studies and officially associated genes is small. When comparing all five diseases and their associated genes there is no overlap among any of them, there are however partial overlaps (Figure 2) (Supplementary Materials 1–5).

ED, pathologic neovascularization, cardiovascular pregnancy complications, and PE overlap in *ANGPT2* which encodes Angiopoietin 2, a protein responsible for

**Table 1:** Top five PE related genes.

| Gene | Full gene name | DSIg | Example animal model strain | Application |
|---|---|---|---|---|
| *ZNF295-AS1* | ZNF295 antisense RNA 1 | 1.000 | NA | NA |
| *ASB4* | Ankyrin repeat and SOCS box containing 4 | 0.931 | C57BL/6-Asb4em1Smoc<br>C57BL/6J-Asb4em1cyagen | Metabolism (Li et al. 2010; Yazdi et al. 2015)<br>PE (Townley-Tilson et al. 2014) |
| *FLT1P1* | FLT1 pseudogene 1 | 0.931 | Prox1-GFP/Flt1-DsRed | PE (Maynard, 2003)<br>Angiogenesis (Zhong et al. 2017) |
| *KIR3DL3* | Killer cell immunoglobulin like receptor, three Ig domains and long cytoplasmic tail 3 | 0.861 | NA | NA |
| *GCM1* | Glial cells missing transcription factor 1 | 0.839 | STOCKTg(Gcm1-cre)1Chrn/J | Reproductive biology research (Woods et al. 2018) |

These genes are sorted by disease specificity index (DSIg), available animal models based on overexpression or KO of gene of interest, and their field of application. ZNF295-AS1, ASB4, FLT1P1, KIR3DL3, and GCM1 show the highest disease specificity score. Animal models only exist for ASB4, FLT1P1, and GCM1 overexpression or knockout. The ASB4 and FL1P1 models have been successfully established in PE research.

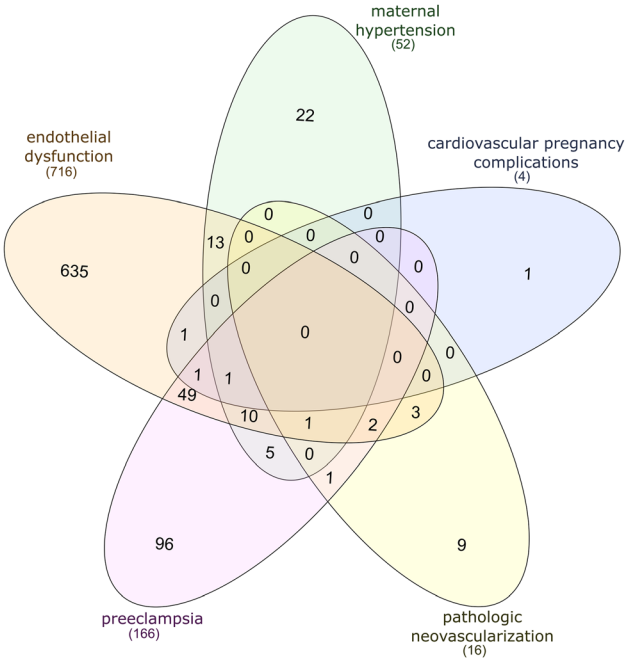



**Figure 2:** Venn diagram of overlapping genes associated with ED, PE, pathologic neovascularization, cardiovascular pregnancy complications, or maternal hypertension.
The total number of gene-disease associations is shown below the disease name.

vascular remodelling (Tabruyn et al. 2010). ED, maternal hypertension, cardiovascular pregnancy complications and PE are all associated with *TNF* (Tumour Necrosis Factor), which is responsible for apoptosis modulation (Polunovsky et al. 1994). PE, ED, and pathologic neovascularization are associated with *CLU* and *VEGFA*, encoding Clustering and Vascular Endothelial Growth Factor A, respectively. Clustering is also implicated in apoptosis (Cunin et al. 2016). For the sake of comparison, the phenotypes were considered separate pathologies. Considering the still obscure pathoetiology of PE, there might be more overlap, which has yet to be discovered. Notably, most of the overlap in gene-disease associations occurs for genes associated with either vascularization or apoptosis.

## Mitochondrial function in pregnancy and preeclampsia

Mitochondria are intracellular organelles that have a plethora of functions in the cell and are nicknamed the powerhouse of the cell. Their primary functions are the production of ATP, the energy currency of the cell and to regulate cellular metabolism (Kühlbrandt 2015). The mammalian mitochondrial electron transport chain (ETC) consists of the inner membrane-embedded complexes I-IV, and the electron transporters cytochrome *c* and ubiquinone (Zhao et al. 2019). The oxidative phosphorylation system (OXPHOS) complex comprises the ETC and complex V. Each complex consists of catalytic activity performing core proteins and a large number of subunits supporting assembly, regulation and stability. Complex assembly is further supported by OXPHOS factors which are encoded by both mitochondrial and nuclear DNA (Signes and Fernandez-Vizarra 2018). The ETC generates a proton gradient across the mitochondrial inner membrane, which is used by complex V to produce ATP. Some electrons are transferred to $O_2$, causing the generation of reactive oxygen species (ROS) (Zhao 2019). Complex I is also involved in superoxide production via paraquat (Cochemé and Murphy 2008).

Dysfunctional mitochondria have been shown to contribute to a wide range of disorders, such as hypertension, diabetes, cancers, and neurodegenerative disorders (DiMauro et al. 2010; Ježek et al. 2010; Stark and Roden 2007). Mitochondria have also come into focus regarding their role in PE development. Given their wide range of functions, deregulated mitochondria may affect placentation on several levels. Defective mitochondria, specifically, β-cell mitochondrial alteration, are associated with insufficiency in the placenta (Rodríguez-Rodríguez et al. 2018). Changes in structure and activity of mitochondria have an impact on the electron transport chain in the inner membrane and subsequently on ATP production. Cytochrome *c* oxidase, a marker enzyme of the mitochondrial inner membrane is reduced in preeclamptic placentas (He et al. 2004). Another hypothesis claims that defects in trophoblastic mitochondria may be partially responsible in the pathophysiological chain reaction of PE (Torbergsen et al. 1989).

Oxidative stress is considered to occur as a consequence of an overproduction of reactive oxygen and nitrogen species (RONS), or as a result of insufficient supply of antioxidants needed to dispose of the free radicals and the buffering capacity of the cellular defence mechanisms is exceeded. This excess of free radicals combined with a diminution of antioxidants is believed to play an important role in the pathogenesis underlying PE. In a longitudinal study, Toescu et al. investigated oxidative stress levels at the end of each trimester and 8 weeks postpartum of 17 normotensive pregnancies showing that late gestation was associated with the formation of oxidative stress (Toescu et al. 2002). This and other studies have shown that oxidative stress is expected in healthy

placental development. Nonetheless, increased oxidative stress and reduced placental protective factors have been observed in preeclamptic phenotypes (Rodgers et al. 1988; Wang et al. 2014; Wickens et al. 1981), in both the maternal circulation and the placenta (Cester et al. 1994; Hansson et al. 2015).

Mitochondria are a main source of reactive oxygen species (ROS) and are enriched in polyunsaturated fatty acids, which could be an important source of oxidative stress and lipid peroxidation (Wang 2014). It has been shown that an abnormal increase in lipid peroxidation is upheld by placental mitochondria present in PE. This is due to both an increase in their amount and an increase in their likelihood to oxidation and mitochondrial generation of superoxide leading to the production of an important source of oxidative stress (Wang 1998).

The number of mitochondria can affect the efficiency of mitochondrial function, the interpretation of functional assays. Additionally, the number of mitochondria was shown to be altered in PE (Holland et al. 2018). Pro-fusion mitochondrial dynamin like GTPase (*OPA1*) expression was affected by gestational age and interaction of PE-gestational age (Holland et al. 2017). OPA1 stimulates inner membrane fusion and couples fusion to oxidative phosphorylation.

## The relationship between mitochondrial function, endothelial function and angiogenesis

Studies on murine models of preeclampsia have shown that mitochondria in PE or PE-like phenotypes (models) are dysfunctional. One study (Jiang et al. 2015) looking at a sFlt-1 overexpression model, in which pregnant mice were injected consecutively with sFlt-1 for 8 days, found that placental mitochondria were dysfunctional. They showed that mitochondria in the trophoblast cells in the placenta of sFlt-1 injected mice were swollen and enlarged and that these mitochondria also presented with extensive DNA damage. These sFlt-1 treated mice also exhibited an increase in apoptosis markers. Another study, looking at mice overexpressing STOX1 (Doridot et al. 2014) demonstrated an increase in mitochondrial mass and mutations in genes that are involved in mitochondrial function (Doridot 2014; Smith et al. 2021). It has further been shown that the damage of placental vessels caused by increased sFLT-1, eventually leads to hypoxia and oxidative stress of trophoblasts, leading to further secretion of sFlt-1 (Jiang 2015; Y et al. 2020). Indeed, oxidative stress induced advanced oxidation protein products treatment of trophoblasts, also leads to increased sFLT-1 expression and consequently systemic and local ED, potentially creating a vicious cycle of oxidative stress, sFLT-1 expression, and ED (Huanga et al. 2013; Murphy et al. 2013; Wang et al. 2017).

The mechanism by which sFlt-1 leads to ED is still poorly studied. As described, mitochondria play a central role in apoptosis signalling. Previous studies have shown that ED can be brought on by the cells undergoing apoptosis through the mitochondria-dependent apoptosis pathway, initiated by caspase-9. Zhai et al. have shown that increased sFlt-1 can activate caspase-9 and caspase-3, indicating sFlt-1 mediated apoptosis (Zhai et al. 2020).

Moreover, research has shown that ED, which in PE leads to angiogenic imbalance and to an insufficient oxygen supply and nitric oxide availability, is caused by oxidative stress (Sena et al. 2018; Smith 2021). Given the link between oxidative stress and mitochondria, some researchers have hypothesized that antecedent mitochondrial factors could engender the development of endothelial cell dysfunction. Several of these studies (Mccarthy and Kenny 2016; Sena 2018; Tenório et al. 2018) showed that in different tissues or organelles, be it plasma, endothelial cells or antioxidants, that have been exposed to PE all showed decreased mitochondrial function and increased ROS production. Once again, the correlation of increased ROS production with decreased mitochondrial function illustrates the role oxidative stress plays in both mitochondrial dysfunction and PE.

It has also been shown that oxidative stress contributes to sFlt-1 induced vascular dysfunction (Bridges et al. 2009) and that advanced oxidation protein products, which are toxins created during oxidative stress, enhance sFlt-1 expression in trophoblasts. The exact mechanism, however, is poorly understood. The current hypothesis is that oxidative stress induces the NADPH oxidase dependent pathway, which leads to an increase in sFLT-1 expression (Huanga 2013). Additionally, it has been shown that dysfunctional placentas, such as an ischaemic placenta, release sFlt-1 as well as other receptors (i.e. angiotensin 2) and cytokines which in turn cause endothelial dysfunction (Lamarca 2012). Endothelial dysfunction can manifest in the form of elevated circulating endothelin (ET-1), RONS and increased vascular sensitivity to angiotensin 2 (Lamarca 2012).

Oxidative is the major cause of ED. However, other factors, such as microparticles have been associated with ED. Microparticles are a subgroup of extracellular vesicles and can be produced by various cell types and can have both protective and pathologic effects. Of particular interest in the context of PE are endothelial microparticles

(EMPs) and trophoblast derived microparticles. The production of EMPs occurs under healthy conditions, but is increased under stress conditions, such as apoptosis (Jimenez et al. 2003) and serve as a surrogate marker of ED (Brodsky et al. 2004). Increased EMPs have been shown in cardiovascular diseases and inflammatory disorders, and PE. EMP were shown to lead to endothelial activation and to stimulate the expression of pro-inflammatory cytokines, such as IL-6 and of adhesion molecules, such as VCAM-1 and E-selectin (Morel et al. 2009). High levels of EMPs can inhibit angiogenesis, by impairing endothelial function, diminishing both nitric oxide production and increasing mitochondrial ROS production (Hu et al. 2018; Mezentsev et al. 2005). EMPs can further lead to thrombus formation, reducing uteroplacental blood flow, and consequently directly exacerbating placental hypoxia (Aharon and Brenner 2011) and indirectly by causing proteinuria and dilation failure in the kidney, generating additional hypertension and vasoconstriction in other organs (Roumeliotis et al. 2020).

As a result of an increase in trophoblast apoptosis, syncytiotrophoblast derived microparticles have been studied as possible biomarkers for PE. Syncytiotrophoblasts have been shown to produce more microparticles in PE, the exact pathway by which this leads to apoptosis is poorly understood (Levine et al. 2020). It is still debated which of soluble factors or microparticles play the more important role in angiogenesis deregulation. There is however some evidence supporting a focus on soluble factors (O'Brien et al. 2017).

Given the extensive effects microparticles can have, these have been investigated for therapeutic approaches. Native, or engineered extracellular vesicles, microparticles, and their membranes may be used to manipulate their biological effect (Kao and Papoutsakis 2019).

PE is a risk factor for developing cardiovascular diseases later in life (Thilaganathan and Kalafat 2019). The vascular endothelium is critical for maintaining vascular homeostasis and function. ED is the common feature for cardiovascular disorders including hypertension and diabetes (Varga et al. 2020). The underlying mechanisms of the dysfunctional EC are not fully explored. It has been proposed that cellular senescence, a process which leads to proliferation arrest, is the key driver for cardiovascular diseases (Erusalimsky and Kurz 2006; Erusalimsky 2009; Jia et al. 2019; Marcu et al. 2017; Reicharda and Asosingha 2019). Senescent EC express increased levels of adhesion molecules, VCAM-1 and ICAM-1, and are more susceptible to apoptosis leading them to be more prone to inflammation, atherosclerosis and thrombosis (Erusalimsky 2006; Erusalimsky 2009). Senescence can be initiated by a variety of factors, including oxidative stress, which is also associated with PE (Childs et al. 2018; Zhu et al. 2017). Increased cellular senescence, including endothelial progenitor cell and placental cells, have been observed in PE (Cindrova-Davies et al. 2018; Sugawara et al. 2005). These changes may contribute to ED and later cardiovascular complications in PE.

Evidence suggests that mitochondrial dysfunction plays an important role in activating cell senescence (Vasileiou et al. 2019). It has been demonstrated that EC senescence is mediated by mitochondrial fission and endoplasmic reticulum (ER) stress (Miyao et al. 2020). Interestingly (Sánchez-Aranguren 2014), demonstrated that sFlt-1 induced metabolic perturbations lead to endothelial and mitochondrial dysfunction in PE (Sánchez-Aranguren et al. 2018). In addition, dysregulation of hydrogen sulphide ($H_2S$) pathway resulted in mitochondrial dysfunction and increased sFlt-1 production in ECs (Carolina Sanchez-Aranguren 2020), suggesting a feedback loop between mitochondrial dysfunction and angiogenic imbalance in PE. We propose that EC senescence triggered by increased sFlt-1 and mitochondrial dysfunction contribute to ED and long-term cardiovascular complications associated with PE. More studies are required to provide comprehensive insight into the relationships between mitochondrial dysfunction and the long-term cardiovascular burden.

## Experimental methods to assess mitochondrial function

There are several experimental methods available to assess mitochondrial function. Due to the wide range of mitochondrial functions, these should be used in conjunction to gain maximal information about the system (Table 2).

Oxygen Consumption Rate (OCR) is one of the primary methods used to analyse mitochondrial respiratory activity and mitochondrial dysfunction (Brand and Nicholls 2011; Connolly et al. 2018; Dranka et al. 2011). Using sequential addition of mitochondrial respiratory inhibitors, specific respiratory states can be isolated and modularly assessed. OCR measurements can further be coupled with the extracellular acidification rate (ECAR) to calculate lactate release and anaerobic glycolysis (Mookerjee et al. 2017).

The mitochondrial membrane potential ($\Delta\psi m$), established as the difference in electrical potential between mitochondrial matrix and cytosol can be used as an indicator for the proton-motive force ($\Delta p$). The mitochondrial membrane potential influences ionic transport and more

**Table 2:** The most common *in vivo* and *in vitro* measuring methods for mitochondrial function.

| Method | Input | Output | Reference |
|---|---|---|---|
| Measurement of the oxygen consumption rate (OCR) | *In vitro*<br>Mitochondrial respiratory inhibitors | – Basal respiration<br>– $H^+$ leak<br>– ATP turnover<br>– Maximal respiration<br>– Spare respiratory capacity<br>– Non-mitochondrial $O_2$ consumption | (Brand 2011; Dranka 2011) |
| Extracellular acidification rate (ECAR) | *In vitro*<br>Coupled with OCR | – Anaerobic glycolysis | (Mookerjee, 2017) |
| Mitochondrial membrane potential | *In vitro*<br>Coupled with OCR<br>– Time-series TMRM fluorescence measurement | Indicator for<br>– Respiration<br>– Ionic fluxes across the mitochondrial membrane<br>– ATP synthesis<br>– Apoptosis<br>– ROS or $Ca^{2+}$ mediated cell injury | (Ward, 2007) |
| Mitochondrial NAD(P)H | Coupled with mitochondrial membrane potential<br>– Endogenous auto fluorescence<br>– High-performance liquid chromatography<br>– Enzymatic assays<br>– Labelled spectrometry | Information on<br>– TCA cycle activity<br>– Respiratory chain activity<br>– $NAD^+$ consumption | (Brand, 2011) |
| Respiratory complex activity | – Western Blot<br>– Histochemical staining | – Respiratory complex subunit expression<br>– More useful when variation is already known | (Havlíčková Karbanová 2012; Sipos et al. 2003) |
| Mitochondrial ATP | – Fluorescent reporters<br>– Bioluminescence-based probes | – ATP quantity | (Imamura et al. 2009; Morciano et al. 2017) |
| Mitochondrial $Ca^{2+}$ | – Fluorescent reporters<br>– Two photon microscopy | – $Ca^{2+}$ quantity | (Pozzan and Rudolf, 2009) |
| Mitochondrial pH | – Fluorescent reporters | – pH<br>– In combination with mitochondrial membrane potential and proton-motive force | (Santo-Domingo and Demaurex, 2012) |
| ROS | – Redox-sensitive fluorophores<br>– Fluorescent reporters<br>– Electron paramagnetic resonance spectroscopy<br>– HPLC | – ROS quantity | (Dickinson et al. 2010; Woolley et al. 2013) |
| Mitochondrial morphology | – Cationic probes<br>– MitoTrackers<br>– Fluorescent reporters<br>– Transmission electron microscopy | – Number of mitochondria<br>– Mitochondrial size<br>– Indicator for fusion/fission events | (Barsoum, 2006) |
| *In vivo* measurements of $Ca^{2+}$, pH, redox state | – Genetically encoded fluorescent reporters<br>– Two photon microscopy<br>– Nuclear magnetic resonance | – $Ca^{2+}$ pH<br>– Redox state | (Breckwoldt 2014; Tada 2014) |
| MRS | Requires continuous monitoring | Intracellular oxygenation | (Richardson, 2002) |
| NIRS | Requires continuous monitoring | Oxygen consumption | (Hamaoka, 1996) |

The name, necessary input, output, and example references are shown. As indicated most methods are used in conjunction with at least one additional method to maximise interpretability.

specifically $Ca^{2+}$ transport across the mitochondrial membrane. Fluctuations within it can indicate disrupted respiration, ionic fluxes across the mitochondrial membrane, or ATP synthesis (Connolly 2018; Ward et al. 2007). More pronounced mitochondrial membrane polarization can correlate with apoptosis, $Ca^{2+}$-mediated, or ROS mediated cell injury (Brand 2011).

The pyridine nucleotides $NAD^+$ and NADH and their phosphorylated counterparts $NADP^+$ and NADPH are central to maintenance of redox status and mitochondrial energy metabolism. Electrons from NADH and NADPH reactions can be transferred to further available oxygen molecules and to generate superoxides (ROS subspecies) (Blacker and Duchen 2016). When combined with mitochondrial membrane potential, decreased NAD(P)H measurements can be indicative of reduced TCA cycles and enhanced respiratory chain activity, or increased $NAD^+$ consumption (Brand 2011; Connolly 2018). When a variation in mitochondrial respiratory complexes is detected, a careful study of actual respiratory complex activity and subunit expression is warranted, as compensatory mechanisms have been shown in other studies (Havlíčková Karbanová et al. 2012).

One of the most studied functions of mitochondria is its ATP generation to sustain cellular energy requirements, which is a classic indicator of mitochondrial function (Murphy et al. 2016). $Ca^{2++}$, on the other hand, regulates components of the respiratory chain, substrate import, and ATP synthesis. Mitochondria are key players in $Ca^{2+}$ signalling and homeostasis, by up taking large amounts of it along the mitochondrial membrane gradient and the $Ca^{2+}$ uniporter complex, thus serving as buffers in excitotoxic conditions (Qiu et al. 2013) that may otherwise lead to RONS production and mitochondrial permeability (Pivovarova and Andrews 2010). Measuring ATP/ADP and $Ca^{2+}$ requires the additional confirmation of mitochondrial targeting or co-localization. Although the quantity of said biomarkers can be indicative of mitochondrial function, they do not carry causal information. Knowledge about mitochondrial matrix pH and pH difference between the mitochondrial matrix and cytosol is required to correct experiments involving fluorescent reporters and to correctly calculate the proton-motive force ($\Delta p$). Mitochondrial pH changes often indicate energy metabolism fluctuations as it influences the flux of metabolites across the mitochondrial membrane.

Mitochondria are the primary consumers of oxygen within the cell and have a prominent role in ROS metabolism. ROS can be generated within mitochondria via electron leaks, TCA cycle enzymes, increased respiration and hyperpolarization of the mitochondrial membrane, play a major role in redox-dependent signalling, are important second messengers, but can also contribute to oxidative stress and cell death (Abramov et al. 2007; Bindokas et al. 1996; Lin and Beal 2006; Lopez-Fabuel et al. 2016; Murphy 2009). Mitochondria undergo constant remodelling through fusion and fission, which can impact mitochondrial parameters (Barsoum et al. 2006; Galloway et al. 2012).

*In vivo* measurements are possible, but remain challenging. Genetically engineered imaging tools, such as fluorescent reporters for $Ca^{2+}$, pH, and redox state, can be integrated. The photon imaging and confocal microscopies of the living animal require surgery to access the area of interest (Breckwoldt et al. 2014; Park et al. 2013; Tada et al. 2014). Existing techniques have been developed for neurobiological studies and accordingly may be difficult to adjust to other tissues. MRS and NIRS are promising non-invasive techniques that could be used to measure *in vivo* oxygen consumption. As the reduction of oxygen is the required precursor event to ATP generation, mitochondrial function can be indirectly assessed via the rate of oxygen consumption. NIRS measures optical absorption changes between oxy- and deoxy-haem groups and is thus an indicator for the balance between oxygen supply and consumption (Hamaoka et al. 1996; Richardson et al. 2002).

To adequately make inferences on mitochondrial function and causal relationships, the combination of multiple methods is recommended (Connolly 2018).

## Computational mitochondria models

Mitochondria are responsible for a diversity of functional roles, which result from the complexity of the interaction between mitochondrial components, ranging from ATP production and cellular metabolism regulation, to apoptosis (Galluzzi et al. 2012; Murphy 2016). These often cannot be measured directly using current experimental techniques. Thus, computational modelling of mitochondria became an important tool to study mitochondrial function (Jafri and Kumar 2014). High-throughput methods and large heterogeneous datasets have become common; however, their integration, analysis, standardization, and interpretation remain challenging. Computational models are mathematical representations of the current state of knowledge, that allow for a build-up on traditional experimental approaches (Brodland 2015) and are used to address the aforementioned challenges.

Contrary to beliefs, computational models cannot replace experiments, they can however test hypotheses, lead to new insights, suggest and refine experimental designs, interpret experiments, and support or refute whether a certain mechanism could or could not lead to the phenotype of interest (Brodland 2015). Computational modelling should be considered a complementary method to laboratory experiments. Not to mention, models are based on laboratory experiments and empirical data, as well as simulations based on computational models. These models can therefore inform and pinpoint the need for new experiments, creating new data to be integrated and ultimately improve the model, making it an iterative process.

Models tend to be specific to a certain question. The complexity of mitochondrial functions led to the creation of various mitochondrial models that are representative of specific mitochondrial functions (Aon and Cortassa 2012; Beard 2005; Zhang et al. 2018; Zhu et al. 2020). The construction of a detailed model encompassing all functions mentioned above, is tempting, however computationally expensive and therefore not useful.

Mitochondrial function has traditionally been studied using thermodynamic models (Pietrobon et al. 1986; Westerhoff et al. 1982). More recent kinetic and thermo-kinetic models improved our understanding of mitochondrial energy metabolism and mitochondrial interaction with cytoplasm and other compartments, i.e. identifying ROS as a depolarization trigger and contributor to metabolic oscillation (Aon 2012). In the case of mitochondria, different models such as mitochondrial swelling, proving that mitochondrial deformation can alleviate osmotic pressure (Makarov et al. 2018); motility, identifying parameters that affect mitochondria positioning to regions of high energy demand (Zhu 2020); apoptosis, stressing the importance of cytochrome *c* in mitochondrial respiration (Huber et al. 2011); bioenergetics, illuminating bioenergetics differences of mitochondria from different tissues (Zhang 2018); and dynamics and dysfunction with respect to ageing (Hoffman et al. 2017) were created.

In general, focus was given to metabolic models, including the tricarboxylic acid cycle (Smith and Robinson 2011) and the respiratory chain (Malik and Czajka 2013), which also focuses on mitochondrial disease research (Beard 2005; Maldonado et al. 2019). Table 3 presents a more thorough overview of existing computational mitochondria models.

In recent years, tools to study mitochondrial function on an integrative level have been developed (Brunk et al. 2018; Rahman et al. 2017; Smith et al. 2017) and are publicly available (Malik-Sheriff et al. 2020). Network-based approaches are particularly promising considering their application in complex molecular interactions and data integrations (Barabási et al. 2011). Molecular interaction maps (MIMs), a systems biology network approach, enables multi-omics integration and the creation of entire disease models. MIMs are representations of molecular processes that can be translated into computational models. It presents a knowledge resource by providing and integrating information about i.e. genes, proteins, and molecules. MIMs can be used for advanced network analyses and drug target identification (Mazein et al. 2018; Serhan et al. 2020).

Popular examples of established network mitochondrial models are Recon3D (Brunk 2018) and Leigh Map (Rahman 2017). Recon3D is a genome scale network including three-dimensional metabolite and protein structure information and can be used to functionally characterize disease – gene variant association and metabolic signatures: It has been successfully used to map mitochondrial trifunctional protein deficiency, SUCLA2-related mtDNA depletion syndrome, and phosphate carrier deficiency data (Maldonado 2019; Sahoo et al. 2012). Leigh Map is a manually curated gene to phenotype map used as a Leigh syndrome diagnostic tool and may be adjusted in the future to study other mitochondrial diseases (Rahman 2017). These examples are specific to mitochondrial diseases. PE, as a complex disease, is not considered a mitochondrial disease, nonetheless mitochondrial dysfunction is hypothesized to play a role. In the case of PE a model that can combined with other models of processes considered to be important.

Intracellular biological interactions can be represented by various types of mathematical graph structures. In the common MIM biological entities are traditionally visualized as nodes and reactions as edges. Modelling tools like CellDesigner or NewtEditor allow heteropartite graph creation including compartmentalization, both automatic and manual annotation, in both the human and machine readable SBML. Mitochondrial dysfunction can be described using this method as seen in the model our research group is currently developing (Figure 3). This example is a combination of progress description and activity flow. The two are traditionally not used in the same map, do however increase human interpretability with progress descriptions including detailed mechanisms of biomolecular interactions and activity flow describes the flow of information between elements within a map (Touré et al. 2018). The map includes proteins, receptors, chemical entities, phenotypes and functional complexes that have been associated with mitochondrial dysfunction. As the quantity of mitochondria allows inferences on their activity, the map also includes mitochondrial fission and fusion, as well as mitophagy (Burton and Jauniaux 2018).

**Table 3:** Selection of the most popular computation mitochondria models, sorted by year of publication.

| Author | Model description | New insight | Limitations |
|---|---|---|---|
| (Aon, 2012) | – Mitochondrial energetics<br>– ROS scavenging system<br>– ROS activated anion efflux pathway across inner membrane | – ROS triggers synchronized depolarization<br>– Sensitivity of the IMAC channel can allow for the oscillations + oxidative phosphorylation + cardiac myocyte action potential and calcium | Specific to cardiac mitochondria |
| (Beard, 2005) | – 17 differential equations<br>– 16 adjustable parameters,<br>– Estimated by simultaneously fitted model-simulated steady states and thermodynamics | – Quantitative model<br>– Feedback control is the principle metabolic control mechanism in mitochondrial electron transport | Not detailed enough to create mechanistic model, yet |
| (Jafri 2014) | – Experimental data | – Predicts how pH and Mg influence metabolic oscillations<br>– Decreasing extra-mitochondrial pH decreases oscillation frequency<br>– Increase in Mg leads to increase in frequency and amplitude of oscillations extremely high Mg abolishes oscillations | Specific to cardiac mitochondria |
| (Huber, 2011) | – Mitochondrial respiration<br>– Cyt-*c* | – Loss of Cyt-*c* decreased mitochondrial respiration by 95% and depolarised mitochondrial membrane potential<br>– Quantitative and mechanistic explanation of protective role of enhanced glucose utilisation | Cancer specific |
| (Hoffman, 2017) | – UPR<br>– Mitochondrial biogenesis<br>– Autophagy dynamics | Physiological parameter changes in mitochondria due to ageing<br>– Declines $NAD^+$ and ATP<br>– Increase in ROS | Specific to ageing |
| (Connolly, 2018) | – Mitochondrial membrane potential<br>– Oxygen consumption rate | – Decreased mitochondrial respiratory capacity could be explained by impairment of mitochondrial NADH flux<br>– This impairment would lead to lower NADH levels in resting state and after manipulation of the respiratory chain<br>– Impaired glycolytic flux causes mitochondrial dysfunction | Specific to neuronal mitochondria |
| (Zhang, 2018) | – Based on data from isolated lung mitochondria | – Insight into bioenergetics and respiration of lung mitochondria<br>– Difference of lung mitochondria to mitochondria from other tissues | Specific to pulmonary tissue |
| (Makarov, 2018) | – Osmosis<br>– IMM rigidity<br>– Dynamics of ionic/neutral species | – Osmotic pressure can be compensated by the rigidity of the IMM<br>– High amounts of dysfunctional mitochondria can activate mitophagy and apoptosis | Only three ionic species and a simplified respiration mechanism |
| (Zhu, 2020) | – Axonal length<br>– Mitochondria velocity<br>– Length of presynaptic terminal<br>– Time of mitochondrial immobility | – Higher AP frequency leads to a larger number of stationary mitochondria | Specific to neuronal mitochondria |

Each model is described by the new input information included in the model, a simplified summary of new insights derived using the model, and general limitations (AP = action potential, ATP = adenosine triphosphate, Cyt-*c* = cytochrome-*c*, IMAC = mitochondrial inner membrane anion channel, IMM = inner mitochondrial membrane, $NAD^+$/NADH = nicotinamide adenine dinucleotide, ROS = reactive oxygen species, UPR = unfolded protein response).

Several mitochondria specific databases exist, offering data ready to be integrated. MitoCore is a curated constraint-based model for simulating human metabolism, while InterMitoBase provides a network of 2813 proteins and 5883 interactions (Gu et al. 2011; Smith 2017). Smith and Robison 2011 created another comprehensive and annotated model, specific to human heart mitochondria (Smith and Robinson 2016). A full list of available

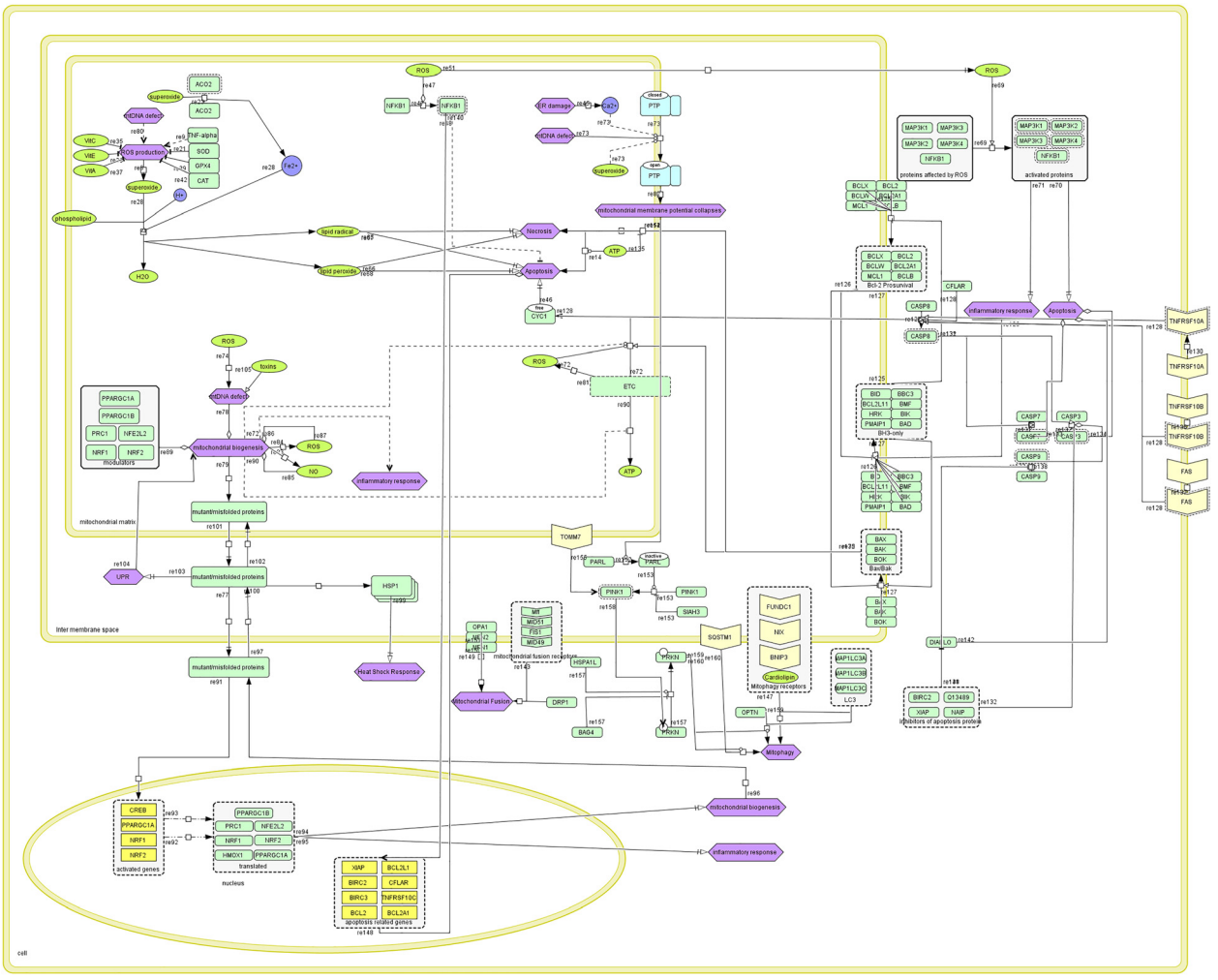



**Figure 3:** Mitochondrial dysfunction progress description and activity flow diagram.
Progress description and activity flow of genes, proteins and molecules associated with mitochondrial dysfunction, and their connection to apoptosis necrosis, mitochondrial fission and fusion, as well as mitophagy in their respective compartment (yellow lines = compartments, green rectangles = proteins, yellow polygon = receptor, bright green ovals = simple molecules, violet hexagon = phenotype, black rectangle = complex, black rectangle dashed line = hypothetical complex, yellow rectangle = gene, blue circle = ion, light blue rectangle = ion channel, arrow = reaction).

databases and tools has been provided by Maldonado et al. (2019). These databases and models have proven useful in various studies and their content could be consolidated into the above-shown network, into a comprehensive molecular interaction model of mitochondrial dysfunction for further analysis and to gain insight into the importance of mitochondrial function on placental dysfunction.

## Mitochondrial interventions

Mitochondrial dysfunction has been associated with multiple diseases such as PE, cancer and neurologic diseases. Thus, a plethora of therapeutic approaches have been invented. Many mitochondrial intervention approaches focus on altering the abundance of pathogenic mutations in mitochondria via heteroplasmic shifting by injection of functional mitochondria, cell therapy, and inhibiting agents. As mitochondria contain multiple copies of their genome they are responsive to this type of therapy (Mercer 2014). Heteroplasmic shifting can be achieved by injecting exogenous mitochondria into cells (King and Attardi 1988; Li et al. 2019). Further, mutated or pathogenic mtDNA replication can be inhibited using peptide nucleic acids (Tanaka et al. 2002). Cell therapy overcomes the challenge of targeting mitochondria *in vivo*. Instead, cells can be extracted, depleted of mitochondria and fused with other cells, containing healthy mitochondria. These

transmitochondrial cell lines have great potential in disease specific models (Bacman and Moraes 2007).

Drugs bound to triphenylphosphonium (TPP), such as the antioxidant MitoQ (Adlam et al. 2005), MitoB (Cochemé et al. 2011), MitoPerox (Prime et al. 2012), and MitoSNO (Prim 2012) can decrease oxidative damage and have been proven beneficial in ischaemic-reperfusion injury (Adlam 2005) and cardiac hypertrophy (Graham et al. 2009) in animal models. Failure of the ubiquitin-proteasome (UPS), which is responsible for the accurate recycling of damaged proteins, has been associated with cardiovascular diseases (Calise and Powell 2013; Herrmann et al. 2010). Both proteasome assembly regulators (Meul et al. 2020) as well as proteasomal inhibitors are currently under investigation as therapeutic interventions (Taylor and Dillin 2011).

Primary mitochondrial disorders in which oxidative phosphorylation is disrupted can further be treated with nutritional interventions. An example of such interventions include micronutrients, signalling modifiers, metabolic agents, and dietary patterns (Ronco and Debiec 2020). More specifically, nutritional supplements include vitamins, antioxidants, minerals, metabolites, herbs, membrane phospholipids, and enzyme inhibitors, as well as cofactors (Kerr 2010) and support metabolic pathways, OXPHOS, which promote mitochondrial biogenesis.

Vitamin B1 enters the cell via thiamine transporters 1 and 2. Supplementation is commonly used to enhance OXPHOS and pyruvate dehydrogenase complex flux. Vitamin B2 (or riboflavin), which is the precursor of flavin adenine dinucleotide, supplementation has shown positive effects in patients with defects of riboflavin transport and mutated flavoprotein dehydrogenase, flavoproteins (Bosch et al. 2011), deficient ETC complex 1 and 2 (Balasubramaniam and Yaplito-Lee 2020; Penn et al. 1992) melas syndrome; riboflavin metabolism. An increase of the precursor of $NAD^+$ and $NADP^+$, vitamin B3, can cause lactic acidosis. This deregulation can be ameliorated by fasting and exercise (Tischner and Wenz 2015). Keep the fire burning: current avenues in the quest of treating mitochondrial disorders. The nervous system penetrating vitamin B9 functions as a B vitamin cofactor required for cellular one-carbon-transfer reactions and has shown positive effects in patients with cerebral folate deficiency.

Antioxidant responses, OXPHOS, and mitochondrial biogenesis can be manipulated and supported using metabolic modifiers, such as α-lipoic acid, L-carnitine, coenzyme Q10, and creatine. α-Lipoic acid (α-LA) is a well studied fatty acid that acts as an enzymatic cofactor in metabolism regulation, mitochondrial biogenesis, and energy production. α-LA also acts as a potent antioxidant further improving mitochondrial performance and contains epigenetic regulatory activity on IL-1B and IL-6 gene expression, which are associated with oxidative stress and inflammation. It's therapeutic benefit has yet to be supported by clinical (Dinicola et al. 2017; Dos Santos et al. 2019). L-carnitine is involved in long-chain fatty acid transport from the cytosol to the mitochondrial matrix and important for β-oxidation, modulation of acyl-CoA/CoA ratio, excretion of toxic acly groups, muscle storage of energy as acetyl-carnitine and additionally functions as a free radical scavenger (Longo et al. 2016). The majority of carnitine is obtained from dietary sources and a common intervention for mitochondrial dysfunction (Oyanagi et al. 2008). The main function of coenzyme Q10 (CoQ10) is as a diffusible electron carrier, transferring electrons from complex I and II to complex III of the ETC, supporting OXPHOS stability. CoQ10 further acts as an effective antioxidant, inhibiting propagation of lipid peroxidation initiation and removing free radicals. Although OXPHOS dysfunction causes an increase of ROS and CoQ10 is a powerful ROS scavenger, the beneficial effects of CoQ10 are still undetermined (Luo et al. 2019). Creatine stores metabolic energy and has been shown to prevent structural and functional damage to mitochondria of oxidative stressed cells (Kazak and Cohen 2020; Ostojic 2018).

Signalling-pathway modulators can be beneficial, when the dysfunctional pathway has been identified. The adaptation of a ketogenic diet is highly controversial. It has been shown that transcripts of TCA cycle, OXPHOS, and glycolysis genes can be increased by adapting a ketogenic diet. During prolonged adoption of ketogenic diet, however, reduced cell respiration, induced cell apoptosis, and decreased mitochondrial biogenesis have been reported (Xu et al. 2021).The bioactive polyphenols resveratrol can correct ETC complex I and IV defects and leads to OXPHOS stabilization when supplemented with metformin. The exact mechanism has yet to be understood (De Oliveira et al. 2016). Nevertheless, a considerable amount of research is still needed to accurately understand the exact processes involved in the metabolism of these therapeutic interventions.

# Circumspections

## Nomenclature

Although PE is a serious pregnancy complication with wide-ranging health, social and economic burdens its exact pathophysiology remains to be revealed. There are several *in vivo, in vitro,* and *in silico* models available to study mitochondrial function. However, specific models

for PE subtypes do not yet exist. Notwithstanding, as studies have shown, in term preeclamptic pregnancies antioxidant activity was increased while in pre-term pre-eclamptic pregnancies antioxidant activity remained unchanged and $H_2O_2$ production, SOD activity and glutathione peroxidase and catalase activity were decreased (Holland 2018).

One of the hindering factors to the progress of the modelling with regards to PE subtypes is the lack of nomenclature definition. For instance, term- and preterm PE, defined as PE occurring after 37 weeks and before 37 weeks of gestation, as well as late onset (LOPE) and early onset PE (EOPE), defined as PE occurring after 34 and before 34 week of gestation (Raymond and Peterson 2011), respectively, present differing phenotypes on a molecular level (Phillips et al. 2010). Not to mention, the definition of PE subtypes itself is controversial.

To facilitate the progress in PE subtype modelling, a global standardization on the PE subtype definition may increase reusability of data and the general interpretability of results. Although, recent studies have shown that a clear separation is necessary as phenotypes differ (Holland 2018), a discordance among the definitions of the current nomenclature still exists, which is undoubtedly to the disadvantage of both animal models and computational models.

Further illustrating the necessity for a clear differentiation in subtype nomenclature in PE, studies showed that when compared to late-onset PE, an increase of oxidative stress and of unfolded protein response (UPR) (protein that is associated with the suppression of non-essential protein synthesis) were found in early-onset PE which could be causing morphological changes. This increase of oxidative stress and UPR explains why growth restriction is often associated with early-onset pre-eclampsia. Despite the wide range of research on the placenta and the different hypotheses on abnormal placentation and how it relates to PE, a significant amount of incertitude remains. Extensive research still needs to be conducted to elucidate this incertitude.

Additionally, as a complex disease PE is often accompanied by other comorbidities, such as hypertension, superimposed with PE, diabetes and asthma (Booker et al. 2019; Czerwinski et al. 2012; Weissgerber and Mudd 2015). These possibly share mechanisms of pathogenesis. Also, the required number of samples for appropriate and robust statistical analysis and interpretation is relatively high, thereby increasing the extent of the analysis needed (Brookhart et al. 2010; Marx et al. 2017).

Computational models cannot replace experiments, nor can they prove whether or not specific mechanisms produce a certain phenotype (Brodland 2015). Nonetheless, network modelling is an established method to study disease-disease associations and diseases in general (Fotouhi et al. 2018). Disease maps are becoming increasingly popular to study complex topics such as Parkinson's disease (Fujita et al. 2014) or inflammation and inflammation resolution (Serhan 2020).

Forasmuch as most animal models of PE are induced by genetic (Collinot et al. 2018), pharmacological (Tam et al. 2011), environmental (Peyronnet et al. 2002), surgical (Crews et al. 2000), or immunological manipulation, resulting in the general preeclamptic phenotype in contrast to PE developing during pregnancy (Gatford et al. 2020). As computational modelling is inherently iterative, these differences may inadvertently be reflected in the model.

## Correlation does not equal causation

Overall, the currently available scientific evidence on PE leads to the assumption that the preeclamptic phenotype is caused by a combination of mitochondrial deregulation, subsequent oxidative stress and consequent ED. The conjunction of oxidative stress and tissue damage may cause a breach of the placental barrier and thus create a leak of foetal and placental-derived factors into the maternal circulation. This leakage of foetal material into the maternal blood stream leads to maternal endothelial damage, elevated oxidative stress, and systemic inflammation. This further suggests that the shedding of nanoparticles such as free Haemoglobin, which causes kidney damage, and miRNA may further exacerbate the inflammation, vascular damage of the placenta as well as systemic oxidative stress.

On a molecular level, mitochondria and in particular mitochondrial DNA (mtDNA) are sensitive to ROS damage in proximity to the oxygen generation site of the ETC. MtDNA damage and mtDNA mutation lead to impaired energy production and risk of further electron leakage which in turn causes an increase in oxidative stress. Mitochondrial DNA levels in the maternal circulation play an important role both in PE and as an indicator of placental abruption. An increase in maternal mtDNA levels has been linked to an increase in oxidative stress. This increase is thought to be due to the hypoxic conditions associated with placental insufficiency and thus a diminution of placental blood flow. Hypoxic stress has been shown to increase the biogenesis of mitochondria, this generating more circulating mtDNA. However, unchanged, as well as both increased and decreased levels of mtDNA have been found in the same pathogenesis of pregnancy

through different studies (Lattuada et al. 2008; Lin 2006; Pillai et al. 2016; Qiu et al. 2012).

The potential causative correlation of oxidative stress and PE has long been debated (Agarwal et al. 2015). Some studies suggested that PE arose as a consequence of an excessive amount of oxidative stress, whereas other studies supported that increased oxidative stress occurs as a consequence of PE. It has also been demonstrated that oxidative stress is a great contributor to endothelial senescence, which could contribute to the phenotype. However, the causative correlation is still debated. Oxidative stress, however, also occurs in other pathologies, suggesting oxidative stress may be a side-effect rather than cause of PE. This may explain the contradicting mtDNA levels across studies (He 2004; Noback 1946; Poidatz et al. 2015).

Although we operate under the assumption that the condition underlying PE can be found somewhere in the placenta, there is a competitive hypothesis about the heart being the cause for PE development. This idea is mainly based on the observation of poor long term cardiovascular outcome in preeclamptic women, and angiogenesis associated biomarkers sFlt-1 and vascular endothelial growth factor (PIGF) which may not only affect the placenta but the heart (Thilaganathan 2019). Further experimental and computational study of mitochondrial function in PE using both cardiac and placenta samples with a focus on oxidative stress, sFlt1, and its effects on EC is required to clarify this discrepancy.

## Conclusion

PE is a multifactorial, complex disease, with wide-ranging health, social and economic burdens, nevertheless its exact pathophysiology remains poorly known. The scientific evidence currently available on PE suggests that the preeclamptic phenotype is caused by a combination of mitochondrial deregulation, subsequent oxidative stress and consequent ED.

Many computational mitochondria models and mitochondria dysfunction models exist, 54 and 19, respectively, can be found on bio models (Malik-Sheriff 2020). Placenta specific models, however, are not available. An in-silico model encompassing the highlighted processes of angiogenesis, oxidative stress, ROS production, mitochondrial function and dysfunction, and ED in combination with data derived from cardiac and placenta tissue samples may elucidate the pathogenesis of PE and the interplay between these processes and identify new biomarkers and therapeutic target candidates for PE.

**Acknowledgments:** This publication reflects only the authors' view and that the European Commission and the Research Executive Agency is not responsible for any use that may be made of the information it contains.

**Author contributions:** All the authors have accepted responsibility for the entire content of this submitted manuscript and approved submission.

**Research funding:** J.S. and Y.C. have received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No 765274, project iPLACENTA.

**Conflict of interest statement:** The authors declare no conflicts of interest regarding this article.

## References

Abramov, A.Y., Scorziello, A., and Duchen, M.R. (2007). Three distinct mechanisms generate oxygen free radicals in neurons and contribute to cell death during anoxia and reoxygenation. J. Neurosci. 27: 1129–1138.

Adlam, V., Harrison, J., Porteous, C., James, A., Smith, R., Murphy, M., and Sammut, I. (2005). Targeting an antioxidant to mitochondria decreases cardiac ischemia-reperfusion injury. FASEB (Fed. Am. Soc. Exp. Biol.) J. 19, https://doi.org/10.1096/fj.05-3718com.

Agarwal, A., Tvrda, E., and Mulgund, A. (2015). Oxidative stress in preeclampsia. In: Watson, R.R.W. (Ed.), *The handbook of fertility: nutrition, diet, lifestyle and reproductive health*. Elsevier Inc, pp. 283–290. https://doi.org/10.1016/b978-0-12-800872-0.00026-3.

Aharon, A. and Brenner, B. (2011). Microparticles and pregnancy complications. Thromb. Res. 127(Suppl. 3): S67–S71.

Aon, M.A. and Cortassa, S. (2012). Mitochondrial network energetics in the heart. Wiley Interdiscip. Rev. Syst. Biol. Med. 4: 599–613.

Apicella, C., Ruano, C.S.M., Méhats, C., Miralles, F., and Vaiman, D. (2019). The role of epigenetics in placental development and the etiology of preeclampsia. Int. J. Mol. Sci. 20: 2837.

Bailey, J., Shaw, A., Fischer, R., Ryan, B.J., Kessler, B.M., Mccullagh, J., Wade-Martins, R., Channon, K.M., and Crabtree, M.J. (2017). A novel role for endothelial tetrahydrobiopterin in mitochondrial redox balance. Free Radic. Biol. Med. 104: 214–225.

Balasubramaniam, S. and Yaplito-Lee, J. (2020). Riboflavin metabolism: role in mitochondrial function. J. Transl. Genet. Genom. 4: 285–306.

Barabási, A.L., Gulbahce, N., and Loscalzo, J. (2011). Network medicine: a network-based approach to human disease. Nat. Rev. Genet. 12: 56–68.

Barsoum, M.J., Yuan, H., Gerencser, A.A., Liot, G., Kushnareva, Y., Gräber, S., Kovacs, I., Lee, W.D., and Bossy-Wetzel, E. (2006). Nitric oxide-induced mitochondrial fission is regulated by dynamin-related GTPases in neurons. EMBO J. 25: 3900–3911.

Beard, D.A. (2005). A biophysical model of the mitochondrial respiratory system and oxidative phosphorylation. PLoS Comput. Biol. 1: 0252–0264.

Bellamy, L., Casas, J.-P., Hingorani, A.D., and Williams, D.J. (2007). Pre-eclampsia and risk of cardiovascular disease and cancer in later life: systematic review and meta-analysis. BMJ 335: 974.

Bergmann, A., Ahmad, S., Cudmore, M., Gruber, A.D., Wittschen, P., Lindenmaier, W., Christofori, G., Gross, V., and Weich, H.A. (2010). Reduction of circulating soluble Flt-1 alleviates preeclampsia-like symptoms in a mouse model. J. Cell Mol. Med. 14: 1857–1867.

Bindokas, V.P., Jordán, J., Lee, C.C., and Miller, R.J. (1996). Superoxide production in rat hippocampal neurons: selective imaging with hydroethidine. J. Neurosci. 16: 1324–1336.

Blacker, T.S. and Duchen, M.R. (2016). Investigating mitochondrial redox state using NADH and NADPH autofluorescence. Free Radic. Biol. Med. 100: 53–65.

Booker, W.A., Ananth, C.V., Wright, J.D., Siddiq, Z., D'Alton, M.E., Cleary, K.L., Goffman, D., and Friedman, A.M. (2019). Trends in comorbidity, acuity, and maternal risk associated with preeclampsia across obstetric volume settings. J. Matern. Fetal Neonatal Med. 32: 2680–2687.

Bosch, A.M., Abeling, N.G.G.M., Ijlst, L., Knoester, H., Van Der Pol, W.L., Stroomer, A.E.M., Wanders, R.J., Visser, G., and Waterham, H.R. (2011). Brown-Vialetto-Van Laere and Fazio Londe syndrome is associated with a riboflavin transporter defect mimicking mild MADD: a new inborn error of metabolism with potential treatment. J. Inherit. Metab. Dis. 34: 159–164.

Brand, M.D. and Nicholls, D.G. (2011). Assessing mitochondrial dysfunction in cells. Biochem. J. 435: 297–312.

Breckwoldt, M.O., Pfister, F.M.J., Bradley, P.M., Marinković, P., Williams, P.R., Brill, M.S., Plomer, B., Schmalz, A., and Misgeld, T. (2014). Multiparametric optical analysis of mitochondrial redox signals during neuronal physiology and pathology *in vivo*. Nat. Med. 20: 555–560.

Bridges, J.P., Gilbert, J.S., Colson, D., Gilbert, S.A., Dukes, M.P., Ryan, M.J., and Granger, J.P. (2009). Oxidative stress contributes to soluble fms-like tyrosine kinase-1 induced vascular dysfunction in pregnant rats. Am. J. Hypertens. 22: 564.

Brodland, G.W. (2015). How computational models can help unlock biological systems. Semin. Cell Dev. Biol. 47: 62–73.

Brodsky, S.V., Zhang, F., Nasjletti, A., and Goligorsky, M.S. (2004). Endothelium-derived microparticles impair endothelial function *in vitro*. Am. J. Physiol. Heart Circ. Physiol. 286, https://doi.org/10.1152/ajpheart.01172.2003.

Brookhart, M.A., Stürmer, T., Glynn, R.J., Rassen, J., and Schneeweiss, S. (2010). Confounding control in healthcare database research: challenges and potential approaches. Med. Care 48, https://doi.org/10.1097/MLR.0b013e3181dbebe3.

Brown, M.A., Magee, L.A., Kenny, L.C., Karumanchi, S.A., McCarthy, F.P., Saito, S., Hall, D.R., Warren, C.E., and Ishaku, S. (2018). The hypertensive disorders of pregnancy: ISSHP classification, diagnosis & management recommendations for international practice. Pregnancy Hypertens. 13: 291–310.

Brunk, E., Sahoo, S., Zielinski, D.C., Altunkaya, A., Dräger, A., Mih, N., Gatto, F., Nilsson, A., and Palsson, B.O. (2018). Recon3D enables a three-dimensional view of gene variation in human metabolism. Nat. Biotechnol. 36: 272–281.

Burke, S.D., Barrette, V.F., Bianco, J., Thorne, J.G., Yamada, A.T., Pang, S.C., Adams, M.A., and Croy, B.A. (2010). Spiral arterial remodeling is not essential for normal blood pressure regulation in pregnant mice. Hypertension 55: 729–737.

Burton, G.J. and Jauniaux, E. (2018). Pathophysiology of placental-derived fetal growth restriction. Am. J. Obstet. Gynecol. 218: S745–S761.

Bacman, Sandra R. and Moraes, Carlos T. (2007). Transmitochondrial technology in animal cells. Methods Cell Biol. 80: 503–524.

Calise, J. and Powell, S.R. (2013). The ubiquitin proteasome system and myocardial ischemia. Am. J. Physiol. Heart Circ. Physiol. 304: 337–349.

Carolina Sanchez-Aranguren, L., Ahmad, S., Dias, I.H.K., Alzahrani, F.A., Rezai, H., Wang, K., and Ahmed, A. (2020). Bioenergetic effects of hydrogen sulfide suppress soluble Flt-1 and soluble endoglin in cystathionine gamma-lyase compromised endothelial cells. Nature 10: 1–11.

Carty, D.M. (2012). *Pre-eclampsia ; early prediction and long-term consequences Institute of Cardiovascular and Medical Sciences*. University of Glasgow.

Cester, N., Staffolani, R., Rabini, R.A., Magnanelli, R., Salvolini, E., Galassi, R., Mazzanti, L., and Romanini, C. (1994). Pregnancy induced hypertension: a role for peroxidation in microvillus plasma membranes. Mol. Cell. Biochem. 131: 151–155.

Chaiworapongsa, T., Romero, R., Espinoza, J., Bujold, E., Mee Kim, Y., Gonçalves, L.F., Gomez, R., and Edwin, S. (2004). Evidence supporting a role for blockade of the vascular endothelial growth factor system in the pathophysiology of preeclampsia: young investigator award. Am. J. Obstet. Gynecol. 190: 1541–1547.

Chen, Wang, Huang, H., and Dong, M. (2008). Vascular endothelial growth factor attenuates Nω-Nitro-L-arginine methyl ester-induced preeclampsia-like manifestations in rats. Clin. Exp. Hypertens. 30: 606–615.

Chen, Y.H., Wu, R., Geng, B., Qi, Y.F., Wang, P.P., Yao, W.Z., and Tang, C.S. (2009). Endogenous hydrogen sulfide reduces airway inflammation and remodeling in a rat model of asthma. Cytokine 45: 117–123.

Childs, B.G., Li, H., and Van Deursen, J.M. (2018). Senescent cells: a therapeutic target for cardiovascular disease. J. Clin. Invest. 128: 1217–1228.

Cindrova-Davies, T., Fogarty, N.M.E., Jones, C.J.P., Kingdom, J., and Burton, G.J. (2018). Evidence of oxidative stress-induced senescence in mature, post-mature and pathological human placentas. Placenta 68: 15–22.

Cochemé, H., Quin, C., McQuaker, S., Cabreiro, F., Logan, A., Prime, T., Abakumova, I., Patel, J., and Murphy, M. (2011). Measurement of $H_2O_2$ within living *Drosophila* during aging using a ratiometric mass spectrometry probe targeted to the mitochondrial matrix. Cell Metabol. 13, https://doi.org/10.1016/j.cmet.2011.02.003.

Cochemé, H.M. and Murphy, M.P. (2008). Complex I is the major site of mitochondrial superoxide production by paraquat. J. Biol. Chem. 283: 1786–1798.

Collinot, H., Marchiol, C., Lagoutte, I., Lager, F., Siauve, N., Autret, G., Balvay, D., Renault, G., and Vaiman, D. (2018). Preeclampsia induced by STOX1 overexpression in mice induces intrauterine growth restriction, abnormal ultrasonography and BOLD MRI signatures. J. Hypertens. 36: 1399–1406.

Connolly, N.M.C., Theurey, P., Adam-Vizi, V., Bazan, N.G., Bernardi, P., Bolaños, J.P., Culmsee, C., Dawson, V.L., and Prehn, J.H.M. (2018). Guidelines on experimental methods to assess mitochondrial dysfunction in cellular models of neurodegenerative diseases. Cell Death Differ. 25: 542–572.

Crews, J.K., Herrington, J.N., Granger, J.P., and Khalil, R.A. (2000). Decreased endothelium-dependent vascular relaxation during reduction of uterine perfusion pressure in pregnant rat. Hypertension 35: 367–372.

Cunin, P., Beauvillain, C., Miot, C., Augusto, J.F., Preisser, L., Blanchard, S., Pignon, P., Scotet, M., and Delneste, Y. (2016). Clusterin facilitates apoptotic cell clearance and prevents apoptotic cell-induced autoimmune responses. Cell Death Dis. 7: e2215.

Czerwinski, S., Gollero, J., Qiu, C., Sorensen, T.K., and Williams, M.A. (2012). Migraine-asthma comorbidity and risk of hypertensive disorders of pregnancy. J. Pregnancy 2012, https://doi.org/10.1155/2012/858097.

De Oliveira, M.R., Nabavi, S.F., Manayi, A., Daglia, M., Hajheydari, Z., and Nabavi, S.M. (2016). Resveratrol and the mitochondria: from triggering the intrinsic apoptotic pathway to inducing mitochondrial biogenesis, a mechanistic view. Biochim. Biophys. Acta Gen. Subj. 1860: 727–745.

Dickinson, B.C., Srikun, D., and Chang, C.J. (2010). Mitochondrial-targeted fluorescent probes for reactive oxygen species. Curr. Opin. Chem. Biol. 14: 50–56.

DiMauro, S., Garone, C., and Naini, A. (2010). Metabolic myopathies. Curr. Rheumatol. Rep. 12: 386–393.

Dinicola, S., Proietti, S., Cucina, A., Bizzarri, M., and Fuso, A. (2017). Alpha-lipoic acid downregulates IL-1β and IL-6 by DNA hypermethylation in SK-N-BE neuroblastoma cells. Antioxidants 6, https://doi.org/10.3390/antiox6040074.

Doridot, L., Châtre, L., Ducat, A., Vilotte, J.L., Lombès, A., Méhats, C., Barbaux, S., Calicchio, R., and Vaiman, D. (2014). Nitroso-redox balance and mitochondrial homeostasis are regulated by STOX1, a pre-eclampsia-associated gene. Antioxidants Redox Signal. 21: 819–834.

Dranka, B.P., Benavides, G.A., Diers, A.R., Giordano, S., Zelickson, B.R., Reily, C., Zou, L., Chatham, J.C., and Darley-Usmar, V.M. (2011). Assessing bioenergetic function in response to oxidative stress by metabolic profiling. Free Radic. Biol. Med. 51: 1621–1635.

Duley, L. (2009). The global impact of pre-eclampsia and eclampsia. Semin. Perinatol. 33: 130–137.

English, F.A., Kenny, L.C., and McCarthy, F.P. (2015). Risk factors and effective management of preeclampsia. Integrated Blood Pres. Contr. 8: 7–12.

Erusalimsky, J.D. and Kurz, D.J. (2006). Endothelial cell senescence. Handb. Exp. Pharmacol. 176(Part 2): 213–248.

Erusalimsky, Jorge D. (2009). Vascular endothelial senescence: from mechanisms to pathophysiology. J. Appl. Physiol. 106: 326–332.

Evans, C.S., Gooch, L., Flotta, D., Lykins, D., Powers, R.W., Landsittel, D., Roberts, J.M., and Shroff, S.G. (2011). Cardiovascular system during the postpartum state in women with a history of preeclampsia. Hypertension 58: 57–62.

Forsythe, J.A., Jiang, B.H., Iyer, N.V., Agani, F., Leung, S.W., Koos, R.D., and Semenza, G.L. (1996). Activation of vascular endothelial growth factor gene transcription by hypoxia-inducible factor 1. Mol. Cell Biol. 16: 4604–4613.

Fotouhi, B., Momeni, N., Riolo, M.A., and Buckeridge, D.L. (2018). Statistical methods for constructing disease comorbidity networks from longitudinal inpatient data. Appl. Netw. Sci. 3, https://doi.org/10.1007/s41109-018-0101-4.

Fujita, K.A., Ostaszewski, M., Matsuoka, Y., Ghosh, S., Glaab, E., Trefois, C., Crespo, I., Perumal, T.M., and Balling, R. (2014). Integrating pathways of Parkinson's disease in a molecular interaction map. Mol. Neurobiol. 49: 88–102.

Galloway, C.A., Lee, H., and Yoon, Y. (2012). Mitochondrial morphology-emerging role in bioenergetics. Free Radic. Biol. Med. 53: 2218–2228.

Galluzzi, L., Kepp, O., Trojel-Hansen, C., and Kroemer, G. (2012). Mitochondrial control of cellular life, stress, and death. Circ. Res. 111: 1198–1207.

Gatford, K.L., Andraweera, P.H., Roberts, C.T., and Care, A.S. (2020). Animal models of preeclampsia: causes, consequences, and interventions. Hypertension 5: 1363–1381.

George, E.M., Liu, H., Robinson, G.G., Mahdi, F., Perkins, E., and Bidwell, G.L. (2015). Growth factor purification and delivery systems (PADS) for therapeutic angiogenesis. Vasc. Cell 7: 8.

Gilbert, J.S., Babcock, S.A., and Granger, J.P. (2007). Hypertension produced by reduced uterine perfusion in pregnant rats is associated with increased soluble fms-like tyrosine kinase-1 expression. Pregnancy Hypertens. 50: 1142–1147.

Graham, D., Huynh, N., Hamilton, C., Beattie, E., Smith, R., Cochemé, H., Murphy, M., and Dominiczak, A. (2009). Mitochondria-targeted antioxidant MitoQ10 improves endothelial function and attenuates cardiac hypertrophy. Hypertension 54, https://doi.org/10.1161/HYPERTENSIONAHA.109.130351.

Gu, Z., Li, J., Gao, S., Gong, M., Wang, J., Xu, H., Zhang, C., and Wang, J. (2011). InterMitoBase: an annotated database and analysis platform of protein-protein interactions for human mitochondria. BMC Genom. 12, https://doi.org/10.1186/1471-2164-12-335.

Hamaoka, T., Iwane, H., Shimomitsu, T., Katsumura, T., Murase, N., Nishio, S., Osada, T., Kurosawa, Y., and Chance, B. (1996). Noninvasive measures of oxidative metabolism on working human muscles by near-infrared spectroscopy. J. Appl. Physiol. 81: 1410–1417.

Hansson, S.R., Nääv, Å., and Erlandsson, L. (2015). Oxidative stress in preeclampsia and the role of free fetal hemoglobin. Front. Physiol. 6: 516.

Havlíčková Karbanová, V., Čížková Vrbacká, A., Hejzlarová, K., Nůsková, H., Stránecký, V., Potocká, A., Kmoch, S., and Houštěk, J. (2012). Compensatory upregulation of respiratory chain complexes III and IV in isolated deficiency of ATP synthase due to TMEM70 mutation. Biochim. Biophys. Acta Bioenerg. 1817: 1037–1043.

He, L., Wang, and Sun, Y. (2004). Reduced amount of cytochrome c oxidase subunit I messenger RNA in placentas from pregnancies complicated by preeclampsia. Acta Obstet. Gynecol. Scand. 83: 144–148.

Herrmann, J., Lerman, L.O., and Lerman, A. (2010). On to the road to degradation: atherosclerosis and the proteasome. Cardiovasc. Res. 85: 291–302.

Hertig, A., Berkane, N., Lefevre, G., Toumi, K., Marti, H.-P., Capeau, J., Uzan, S., and Rondeau, E. (2004). Maternal serum sFlt1 concentration is an early and reliable predictive marker of preeclampsia. Clin. Chem. 50: 1702–1703.

Hoffman, T.E., Barnett, K.J., Wallis, L., and Hanneman, W.H. (2017). A multimethod computational simulation approach for investigating mitochondrial dynamics and dysfunction in degenerative aging. Aging Cell 16: 1244–1255.

Holland, O., Cuffe, J.S.M., Dekker Nitert, M., Callaway, L., Kwan Cheung, K.A., Radenkovic, F., and Perkins, A.V. (2018). Placental mitochondrial adaptations in preeclampsia associated with progression to term delivery. Cell Death Dis. 9: 1–13.

Holland, O., Dekker Nitert, M., Gallo, L.A., Vejzovic, M., Fisher, J.J., and Perkins, A.V. (2017). Review: placental mitochondrial function and structure in gestational disorders. Placenta 54: 2–9.

Hu, Y., Yan, R., Zhang, C., Zhou, Z., Liu, M., Wang, C., Zhang, H., Dong, L., and Wu, Q. (2018). High-mobility group box 1 from hypoxic trophoblasts promotes endothelial microparticle production and thrombophilia in preeclampsia. Arterioscler. Thromb. Vasc. Biol. 38: 1381–1391.

Huanga, Q.T., Wanga, S.S., Zhanga, M., Huanga, L.P., Tianb, J.W., Yua, Y.H., Wanga, Z.J., and Zhong, M. (2013). Advanced oxidation protein products enhances soluble Fms-like tyrosine kinase 1 expression in trophoblasts: a possible link between oxidative stress and preeclampsia. Placenta 34: 949–952.

Huber, H.J., Dussmann, H., Kilbride, S.M., Rehm, M., and Prehn, J.H.M. (2011). Glucose metabolism determines resistance of cancer cells to bioenergetic crisis after cytochrome-c release. Mol. Syst. Biol. 2011: 470.

Imamura, H., Huynh Nhat, K.P., Togawa, H., Saito, K., Iino, R., Kato-Yamada, Y., Nagai, T., and Noji, H. (2009). Visualization of ATP levels inside single living cells with fluorescence resonance energy transfer-based genetically encoded indicators. Proc. Natl. Acad. Sci. U.S.A. 106: 15651–15656.

Jafri, M.S. and Kumar, R. (2014). Modeling mitochondrial function and its role in disease. Prog. Mol. Biol. Transl. Sci. 123: 103–125.

James, J.L., Chamley, L.W., and Clark, A.R. (2017). Feeding your baby in utero: how the uteroplacental circulation impacts pregnancy. Physiology 32: 234–245.

Ježek, P., Plecitá-Hlavatá, L., Smolková, K., and Rossignol, R. (2010). Distinctions and similarities of cell bioenergetics and the role of mitochondria in hypoxia, cancer, and embryonic development. Int. J. Biochem. Cell Biol. 42: 604–622.

Jia, G., Aroor, A.R., Jia, C., and Sowers, J.R. (2019). Endothelial cell senescence in aging-related vascular dysfunction. Biochim. Biophys. Acta (BBA) - Mol. Basis Dis. 1865: 1802–1809.

Jiang, Z., Zou, Y., Ge, Z., Zuo, Q., Huang, S.Y., and Sun, L. (2015). A role of sFlt-1 in oxidative stress and apoptosis in human and mouse pre-eclamptic trophoblasts. Biol. Reprod. 93, https://doi.org/10.1095/biolreprod.114.126227.

Jimenez, J.J., Jy, W., Mauro, L.M., Soderland, C., Horstman, L.L., and Ahn, Y.S. (2003). Endothelial cells release phenotypically and quantitatively distinct microparticles in activation and apoptosis. Thromb. Res. 109: 175–180.

Kao and Papoutsakis, E.T. (2019). Extracellular vesicles: exosomes, microparticles, their parts, and their targets to enable their biomanufacturing and clinical applications. Curr. Opin. Biotechnol. 60: 89–98.

Kazak, L. and Cohen, P. (2020). Creatine metabolism: energy homeostasis, immunity and cancer biology. Nat. Rev. Endocrinol. 16: 421–436.

Kerr, D. (2010). Treatment of mitochondrial electron transport chain disorders: a review of clinical trials over the past decade. Mol. Genet. Metabol. 99, https://doi.org/10.1016/j.ymgme.2009.11.005.

Khan, K.S., Wojdyla, D., Say, L., Gülmezoglu, A.M., and Van Look, P.F. (2006). WHO analysis of causes of maternal death: a systematic review. Lancet 367: 1066–1074.

King, M.P., and Attardi, G. (1988). Injection of mitochondria into human cells leads to a rapid replacement of the endogenous mitochondrial DNA. Cell 52: 811–819.

Koga, K., Osuga, Y., Yoshino, O., Hirota, Y., Ruimeng, X., Hirata, T., Takeda, S., Yano, T., and Taketani, Y. (2003). Elevated serum soluble vascular endothelial growth factor receptor 1 (sVEGFR-1) levels in women with preeclampsia. J. Clin. Endocrinol. Metabol. 88: 2348–2351.

Kühlbrandt, W. (2015). Structure and function of mitochondrial membrane protein complexes. BMC Biol. 13: 89.

Lain, K.Y. and Roberts, J.M. (2002). Contemporary concepts of the pathogenesis and management of preeclampsia. J. Am. Med. Assoc. 287: 3183–3186.

Lamarca, B. (2012). Endothelial dysfunction; an important mediator in the pathophysiology of hypertension during preeclampsia. Minerva Ginecol. 64: 309.

Lambert, G., Brichant, J.F., Hartstein, G., Bonhomme, V., and Dewandre, P.Y. (2014). Preeclampsia : an update. Acta Anaesthesiol. 65: 137–149.

Laresgoiti-Servitje, E. (2013). A leading role for the immune system in the pathophysiology of preeclampsia. J. Leukoc. Biol. 94: 247–257.

Lattuada, D., Colleoni, F., Martinelli, A., Garretto, A., Magni, R., Radaelli, T., and Cetin, I. (2008). Higher mitochondrial DNA content in human IUGR placenta. Placenta 29: 1029–1033.

Levine, L., Habertheuer, A., Ram, C., Korutla, L., Schwartz, N., Hu, R.W., Reddy, S., Freas, A., and Vallabhajosyula, P. (2020). Syncytiotrophoblast extracellular microvesicle profiles in maternal circulation for noninvasive diagnosis of preeclampsia. Sci. Rep. 10: 1–11.

Levine, R.J., Maynard, S.E., Qian, C., Lim, K.-H., England, L.J., Yu, K.F., Schisterman, E.F., Thadhani, R., and contributed, K. (2005). Circulating angiogenic factors and the risk of preeclampsia. Surv. Anesthesiol. 49: 14–15.

Li, F., Xie, Y., Wu, Y., He, M., Yang, M., Fan, Y., Li, X., Qiao, F., and Deng, D. (2019). HSP20 exerts a protective effect on preeclampsia by regulating function of trophoblast cells via Akt pathways. Reprod. Sci. 26: 961–971.

Li, J.Y., Chai, B.X., Zhang, W., Wang, H., and Mulholland, M.W. (2010). Expression of ankyrin repeat and suppressor of cytokine signaling box protein 4 (Asb-4) in proopiomelanocortin neurons of the arcuate nucleus of mice produces a hyperphagic, lean phenotype. Endocrinology 151: 134–142.

Liao, J.K. (2013). Linking endothelial dysfunction with endothelial cell activation. J. Clin. Invest. 123: 540–541.

Lin, M.T. and Beal, M.F. (2006). Mitochondrial dysfunction and oxidative stress in neurodegenerative diseases. Nature 443: 787–795.

Logue, O.C., Fakhri, Chapman, Heather, George, E.M., and Bidwell, G.L. (2017). A maternally sequestered, biopolymer-stabilized vascular endothelial growth factor (VEGF) chimera for treatment of preeclampsia. J. Am. Heart Assoc. 6: 1–15.

Longo, N., Frigeni, M., and Pasquali, M. (2016). Carnitine transport and fatty acid oxidation. Biochim. Biophys. Acta Mol. Cell Res. 1863: 2422–2435.

Lopez-Fabuel, I., Le Douce, J., Logan, A., James, A.M., Bonvento, G., Murphy, M.P., Almeida, A., and Bolaños, J.P. (2016). Complex I assembly into supercomplexes determines differential mitochondrial ROS production in neurons and astrocytes. Proc. Natl. Acad. Sci. U.S.A. 113: 13063–13068.

Luo, K., Yu, J.H., Quan, Y., Shin, Y.J., Lee, K.E., Kim, H.L., Ko, E.J., Chung, B.H., and Yang, C.W. (2019). Therapeutic potential of coenzyme Q10 in mitochondrial dysfunction during tacrolimus-induced beta cell injury. Sci. Rep. 9: 1–12.

Makarov, V., Khmelinskii, I., and Javadov, S. (2018). Computational modeling of *in vitro* swelling of mitochondria: a biophysical approach. Molecules 23: 783.

Makris, A., Thornton, C., Thompson, J., Thomson, S., Martin, R., Ogle, R., Waugh, R., Mckenzie, P., and Hennessy, A. (2007). Uteroplacental ischemia results in proteinuric hypertension and elevated sFLT-1. Kidney Int. 71: 977–984.

Maldonado, E.M., Taha, F., Rahman, J., and Rahman, S. (2019). Systems biology approaches toward understanding primary mitochondrial diseases. Front. Genet. 10: 19.

Malik-Sheriff, R.S., Glont, M., Nguyen, T.V.N., Tiwari, K., Roberts, M.G., Xavier, A., Vu, M.T., Men, J., and Hermjakob, H. (2020). BioModels-15 years of sharing computational models in life science. Nucleic Acids Res. 48: D407–D415.

Malik, A.N. and Czajka, A. (2013). Is mitochondrial DNA content a potential biomarker of mitochondrial dysfunction? Mitochondrion 13: 481–492.

Marcu, R., Zheng, Y., and Hawkins, B.J. (2017). Mitochondria and angiogenesis. Adv. Exp. Med. Biol. 982: 371–406.

Marx, P., Antal, P., Bolgar, B., Bagdy, G., Deakin, B., and Juhasz, G. (2017). Comorbidities in the diseasome are more apparent than real: what Bayesian filtering reveals about the comorbidities of depression. PLoS Comput. Biol. 13: e1005487.

Maynard, S.E., Min, J., Merchan, J., Lim, K.-H., Li, J., Mondal, S., Libermann, T.A., Morgon, J.P., and Karumanchi, S.A. (2003). Excess placental soluble fms-like tyrosine kinase 1 (sFlt1) may contribute to endothelial dysfunction hypertension, and proteinuria in preeclampsia. J. Clin. Invest. 111: 649–658.

Mayrink, J., Costa, M.L., and Cecatti, J.G. (2018). Preeclampsia in 2018: revisiting concepts, physiopathology, and prediction. Sci. World J. 2018, https://doi.org/10.1155/2018/6268276.

Mazein, A., Ostaszewski, M., Kuperstein, I., Watterson, S., Le Novère, N., Lefaudeux, D., De Meulder, B., Pellet, J., and Auffray, C. (2018). Systems medicine disease maps: community-driven comprehensive representation of disease mechanisms. Npj Syst. Biol. Appl. 4: 21.

Mccarthy, C. and Kenny, L.C. (2016). Therapeutically targeting mitochondrial redox signalling alleviates endothelial dysfunction in preeclampsia. Nature 6, https://doi.org/10.1038/srep32683.

Mercer, J. (2014). Mitochondrial bioenergetics and therapeutic intervention in cardiovascular disease. Pharmacol. Therapeut. 141, https://doi.org/10.1016/j.pharmthera.2013.07.011.

Mezentsev, A., Merks, R.M.H., O'Riordan, E., Chen, J., Mendelev, N., Goligorsky, M.S., and Brodsky, S.V. (2005). Endothelial microparticles affect angiogenesis in vitro: role of oxidative stress. Am. J. Physiol. Heart Circ. Physiol. 289: 1106–1114.

Miquerol, L., Langille, B.L., and Nagy, A. (2000). Embryonic development is disrupted by modest increases in vascular endothelial growth factor gene expression. Development 127: 3941–3946.

Miyao, M., Cicalese, S., Kawai, T., Cooper, H.A., Boyer, M.J., Elliott, K.J., Forrester, S.J., Kuroda, R., and Eguchi, S. (2020). Involvement of senescence and mitochondrial fission in endothelial cell pro-inflammatory phenotype induced by angiotensin II. Int. J. Mol. Sci. 21: 1–13.

Mookerjee, S.A., Gerencser, A.A., Nicholls, D.G., and Brand, M.D. (2017). Quantifying intracellular rates of glycolytic and oxidative ATP production and consumption using extracellular flux measurements. J. Biol. Chem. 292: 7189–7207.

Morciano, G., Sarti, A.C., Marchi, S., Missiroli, S., Falzoni, S., Raffaghello, L., Pistoia, V., Giorgi, C., and Pinton, P. (2017). Use of luciferase probes to measure ATP in living cells and animals. Nat. Protoc. 12: 1542–1562.

Morel, O., Pereira, B., Averous, G., Faure, A., Jesel, L., Germain, P., Grunebaum, L., Ohlmann, P., and Toti, F. (2009). Increased levels of procoagulant tissue factor-bearing microparticles within the occluded coronary artery of patients with ST-segment elevation myocardial infarction: role of endothelial damage and leukocyte activation. Atherosclerosis 204, https://doi.org/10.1016/j.atherosclerosis.2008.10.039.

Moser, G., Weiss, G., Gauster, M., Sundl, M., and Huppertz, B. (2015). Evidence from the very beginning: endoglandular trophoblasts penetrate and replace uterine glands *in situ* and *in vitro*. Hum. Reprod. 30: 2747–2757.

Muller, W.A., Weigl, S.A., Deng, X., and Phillips, D.M. (1993). PECAM-1 is required for transendothelial migration of leukocytes. J. Exp. Med. 178: 449–460.

Murphy, E., Ardehali, H., Balaban, R.S., DiLisa, F., Dorn, G.W., Kitsis, R.N., Otsu, K., Ping, P., and Youle, R.J. (2016). Mitochondrial function, biology, and role in disease: a scientific statement from the American heart association. Circ. Res. 118: 1960–1991.

Murphy, M.P. (2009). How mitochondria produce reactive oxygen species. Biochem. J. 417: 1–13.

Murphy, S.R., Babbette, B., Lamarca, D., Parrish, M., Cockrell, K., Granger, J.P., Sr, M., Bb, L., and Granger, K. (2013). Control of soluble fms-like tyrosine-1 (sFlt-1) production response to placental ischemia/hypoxia: role of tumor necrosis factor. Am. Physiol. Soc. 305: R130–R135.

Meul, Thomas, Berschneider, Korbinian, Schmitt, Sabine, Mayr, Christoph H., Mattner, Laura F., Schiller, Herbert B., Yazgili, Ayse S., Wang, Xinyuan, Meiners, Silke, et al. (2020). Mitochondrial regulation of the 26S proteasome. Cell Rep. 32: 108059.

Navarro-Yepes, J., Burns, M., Anandhan, A., Khalimonchuk, O., del RazoLuz, M., Quintanilla-Vega, B., Pappa, A., Panayiotidis, I.M., and Franco, R. (2014). Oxidative stress, redox signaling, and autophagy: cell death versus survival. Antioxidants Redox Signal. 21: 66–85, https://Home.Liebertpub.Com/Ars.

Noback, C.R. (1946). Placentation and angiogenesis in the amnion of a baboon (*Papio papio*). Anat. Rec. 94: 553–567.

O'Brien, M., Baczyk, D., and Kingdom, J.C. (2017). Endothelial dysfunction in severe preeclampsia is mediated by soluble factors, rather than extracellular vesicles. Sci. Rep. 7: 1–12.

Ostojic, S.M. (2018). Plasma creatine as a marker of mitochondrial dysfunction. Med. Hypotheses 113: 52–53.

Oyanagi, E., Yano, H., Kato, Y., Fujita, H., Utsumi, K., and Sasaki, J. (2008). L-Carnitine suppresses oleic acid-induced membrane permeability transition of mitochondria. Cell Biochem. Funct. 26: 778–786.

Papakonstantinou, K., Economou, E., Koupa, E., Babameto, I., Hasiakos, D., and Vitoratos, N. (2011). Antepartum and postpartum maternal plasma levels of E-selectin and VE-cadherin in preeclampsia, gestational proteinuria and gestational hypertension. J. Matern. Fetal Neonatal Med. 24: 1027–1032.

Park, J.M., Josan, S., Grafendorfer, T., Yen, Y.F., Hurd, R.E., Spielman, D.M., and Mayer, D. (2013). Measuring mitochondrial metabolism in rat brain in vivo using MR Spectroscopy of hyperpolarized [2-$^{13}$C]pyruvate. NMR Biomed. 26: 1197–1203.

Penn, A.M.W., Lee, J.W.K., Thuillier, P., Wagner, M., Maclure, K.M., Menard, M.R., Hall, L.D., and Kennaway, N.G. (1992). MELAS syndrome with mitochondrial tRNA Leu(UUR) mutation: correlation of clinical state, nerve conduction, and muscle 31P magnetic resonance spectroscopy during treatment with nicotinamide and riboflavin. Neurology 42: 2147–2152.

Perry, H., Khalil, A., and Thilaganathan, B. (2018). Preeclampsia and the cardiovascular system: an update. Trends Cardiovasc. Med. 28: 505–513.

Peyronnet, J., Dalmaz, Y., Ehrström, M., Mamet, J., Roux, J.C., Pequignot, J.M., Thorén, P.H., and Lagercrantz, H. (2002). Long-lasting adverse effects of prenatal hypoxia on developing autonomic nervous system and cardiovascular parameters in rats. Pflueg. Arch. Eur. J. Physiol. 443: 858–865.

Phillips, J.K., Janowiak, M., Badger, G.J., and Bernstein, I.M. (2010). Evidence for distinct preterm and term phenotypes of preeclampsia. J. Matern. Fetal Neonatal Med. 23: 622–626.

Pietrobon, D., Zoratti, M., Azzone, G.F., and Caplan, S.R. (1986). Intrinsic uncoupling of mitochondrial proton pumps. 2. Modeling studies. Biochemistry 25: 767–775.

Pillai, V.B., Bindu, S., Sharp, W., Fang, Y.H., Kim, G., Gupta, M., Samant, S., and Gupta, M.P. (2016). Sirt3 protects mitochondrial DNA damage and blocks the development of doxorubicin-induced cardiomyopathy in mice. Am. J. Physiol. Heart Circ. Physiol. 310: H962–H972.

Pivovarova, N.B. and Andrews, S.B. (2010). Calcium-dependent mitochondrial function and dysfunction in neurons: Minireview. FEBS J. 277: 3622–3636.

Poidatz, D., Dos Santos, E., Duval, F., Moindjie, H., Serazin, V., Vialard, F., De Mazancourt, P., and Dieudonné, M.N. (2015). Involvement of estrogen-related receptor-γ and mitochondrial content in intrauterine growth restriction and preeclampsia. Fertil. Steril. 104: 483–490.

Polunovsky, V.A., Wendt, C.H., Ingbar, D.H., Peterson, M.S., and Bitterman, P.B. (1994). Induction of endothelial cell apoptosis by TNFα: modulation by inhibitors of protein synthesis. Exp. Cell Res. 214: 584–594.

Possomato-Vieira, J.S. and Khalil, R.A. (2016). Mechanisms of endothelial dysfunction in hypertensive pregnancy and preeclampsia. Adv. Pharmacol. 77: 361–431.

Pozzan, T. and Rudolf, R. (2009). Measurements of mitochondrial calcium *in vivo*. Biochim. Biophys. Acta Bioenerg. 1787: 1317–1323.

Prime, T., Forkink, M., Logan, A., Finichiu, P., McLachlan, J., Li Pun, P., Koopman, W., Larsen, L., and Murphy, M. (2012). A ratiometric fluorescent probe for assessing mitochondrial phospholipid peroxidation within living cells. Free Radical Biol. Med. 53, https://doi.org/10.1016/j.freeradbiomed.2012.05.033.

Qiu, C., Hevner, K., Abetew, D., Sedensky, M., Morgan, P., Enquobahrie, D.A., and Williams, M.A. (2013). Mitochondrial DNA copy number and oxidative DNA damage in placental tissues from gestational diabetes and control pregnancies: a pilot study. Clin. Lab. 59: 655.

Qiu, C., Hevner, K., Enquobahrie, D.A., Williams, M.A., William, M.A., and Williams, M.A. (2012). A case-control study of maternal blood mitochondrial DNA copy number and preeclampsia risk. Int. J. Mol. Epidemiol. Genet. 3: 237–244.

Rahman, J., Noronha, A., Thiele, I., and Rahman, S. (2017). Leigh map: a novel computational diagnostic resource for mitochondrial disease. Ann. Neurol. 81: 9–16.

Raymond, D. and Peterson, E. (2011). A critical review of early-onset and late-onset preeclampsia. Obstet. Gynecol. Surv. 66: 497–506.

Redman, C.W.G. and Sargent, I.L. (2010). Immunology of pre-eclampsia. Am. J. Reprod. Immunol. 63: 534–543.

Rees, D.D., Palmer, R.M.J., Schulz, R., Hodson, H.F., and Moncada, S. (1990). Characterization of three inhibitors of endothelial nitric oxide synthase *in vitro* and *in vivo*. Br. J. Pharmacol. 101: 746–752.

Reicharda, A. and Asosingha, K. (2019). The role of mitochondria in angiogenesis. Mol. Biol. Rep. 46: 1393–1400.

Richardson, R.S., Noyszewski, E.A., Haseler, L.J., Bluml, S., and Frank, L.R. (2002). Evolving techniques for the investigation of muscle bioenergetics and oxygenation. Biochem. Soc. Trans. 30: 232–237.

Roberts, J.M. and Escudero, C. (2012). The placenta in preeclampsia. Pregnancy Hypertens. 2: 72–83.

Roberts, J.M. and Gammill, H.S. (2005). Preeclampsia recent insights preeclampsia: a two-stage disorder. Hypertension 46: 1243–1249.

Roberts, J.M. and Hubel, C.A. (2009). The two stage model of preeclampsia: variations on the theme. Placenta 30: S32–S37.

Roberts, J.M., Taylor, R.N., Musci, T.J., Rodgers, G.M., Hubel, C.A., and McLaughlin, M.K. (1989). Preeclampsia: an endothelial cell disorder. Am. J. Obstet. Gynecol. 161: 1200–1204.

Rodgers, G.M., Taylor, R.N., and Roberts, J.M. (1988). Preeclampsia is associated with a serum factor cytotoxic to human endothelial cells. Am. J. Obstet. Gynecol. 159: 908–914.

Rodríguez-Rodríguez, P., Ramiro-Cortijo, D., Reyes-Hernández, C.G., López de Pablo, A.L., Carmen González, M., and Arribas, S.M. (2018). Implication of oxidative stress in fetal programming of cardiovascular disease. Front. Physiol. 9: 1–13.

Ronco, P. and Debiec, H. (2020). Molecular pathogenesis of membranous nephropathy. Annu. Rev. Pathol. 15, https://doi.org/10.1146/annurev-pathol-020117-043811.

Roumeliotis, S., Mallamaci, F., and Zoccali, C. (2020). Endothelial dysfunction in chronic kidney disease, from biology to clinical outcomes: a 2020 update. J. Clin. Med. 9: 2359.

Dos Santos, S.M., Romeiro, C.F.R., Rodrigues, C.A., Cerqueira, A.R.L., and Monteiro, M.C. (2019). Mitochondrial dysfunction and alpha-lipoic acid: beneficial or harmful in Alzheimer's disease? Oxid. Med. Cell. Longev. 2019, https://doi.org/10.1155/2019/8409329.

Sahoo, S., Franzson, L., Jonsson, J.J., and Thiele, I. (2012). A compendium of inborn errors of metabolism mapped onto the human metabolic network. Mol. Biosyst. 8: 2545–2558.

Samie Omran, A.A. and Mohammed Osman, A.M. (2016). Prediction of preeclampsia with novel biomarkers at second trimester of pregnancy. J. Clin. Cell. Immunol. 7, https://doi.org/10.4172/2155-9899.1000435.

Sánchez-Aranguren, L.C., Espinosa-González, C.T., González-Ortiz, L.M., Sanabria-Barrera, S.M., Riaño-Medina, C.E., Nuñez, A.F., Ahmed, A., Vasquez-Vivar, J., and López, M. (2018). Soluble Fms-like tyrosine kinase-1 alters cellular metabolism and

mitochondrial bioenergetics in preeclampsia. Front. Physiol. 9: 1–13.

Sánchez-Aranguren, L.C., Prada, C.E., Riaño-Medina, C.E., Lopez, M., Pandey, D., and Health, G. (2014). Endothelial dysfunction and preeclampsia: role of oxidative stress Preeclampsia: a disorder of global impact. Front. Physiol. 5, https://doi.org/10.3389/fphys.2014.00372.

Sandoo, A., Veldhuijzen van Zanten, J.J.C., Metsios, G.S., Carroll, D., and Kitas, G.D. (2015). The endothelium and its role in regulating vascular tone. Open Cardiovasc. Med. J. 4: 302–312.

Santo-Domingo, J. and Demaurex, N. (2012). Perspectives on: SGP symposium on mitochondrial physiology and medicine: the renaissance of mitochondrial pH. J. Gen. Physiol. 139: 415–423.

Sena, C.M., Leandro, A., Azul, L., Seiça, R., and Perry, G. (2018). Vascular oxidative stress: impact and therapeutic approaches. Front. Physiol. 9, https://doi.org/10.3389/fphys.2018.01668.

Serhan, C.N., Gupta, S.K., Perretti, M., Godson, C., Brennan, E., Li, Y., Soehnlein, O., Shimizu, T., and Wolkenhauer, O. (2020). The atlas of inflammation resolution (AIR). Mol. Aspect. Med. 74: 100894.

Signes, A. and Fernandez-Vizarra, E. (2018). Assembly of mammalian oxidative phosphorylation complexes I–V and supercomplexes. Essays Biochem. 62: 255–270.

Sipos, I., Tretter, L., and Adam-Vizi, V. (2003). Quantitative relationship between inhibition of respiratory complexes and formation of reactive oxygen species in isolated nerve terminals. J. Neurochem. 84: 112–118.

Smith, A.C. and Robinson, A.J. (2011). A metabolic model of the mitochondrion and its use in modelling diseases of the tricarboxylic acid cycle. BMC Syst. Biol. 5: 102.

Smith, A.C. and Robinson, A.J. (2016). MitoMiner v3.1, an update on the mitochondrial proteomics database. Nucleic Acids Res. 44: D1258–D1261.

Smith, A.C., Eyassu, F., Mazat, J.P., and Robinson, A.J. (2017). MitoCore: a curated constraint-based model for simulating human central metabolism. BMC Syst. Biol. 11: 114.

Smith, A.N., Wang Thomas, D.G., Tatum, R.E., Booz, G.W., and Cunningham, M.W., Jr. (2021). The role of mitochondrial dysfunction in preeclampsia: causative factor or collateral damage? Am. J. Hypertens. 1, https://doi.org/10.1093/ajh/hpab003.

Spradley, F.T., Tan, A.Y., Joo, W.S., Daniels, G., Kussie, P., Ananth Karumanchi, S., and Granger, J.P. (2016). Placental growth factor Administration abolishes placental ischemia-induced hypertension. Hypertension 67: 740–747.

Staff, A.C. (2019). The two-stage placental model of preeclampsia: an update. J. Reprod. Immunol. 134–135: 1–10.

Stark, R. and Roden, M. (2007). Mitochondrial function and endocrine diseases. Eur. J. Clin. Invest. 37: 236–248.

Sugawara, J., Mitsui-Saito, M., Hayashi, C., Hoshiai, T., Senoo, M., Chisaka, H., Yaegashi, N., and Okamura, K. (2005). Decrease and senescence of endothelial progenitor cells in patients with preeclampsia. J. Clin. Endocrinol. Metabol. 90: 5329–5332.

Suzuki, H., Ohkuchi, A., Matsubara, S., Takei, Y., Murakami, M., Shibuya, M., Suzuki, M., and Sato, Y. (2009). Effect of recombinant placental growth factor 2 on hypertension induced by full-length mouse soluble fms-like tyrosine kinase 1 adenoviral vector in pregnant mice. Hypertension 54: 1129–1135.

Sweeney, M. and Foldes, G. (2018). It takes two: endothelial-perivascular cell cross-talk in vascular development and disease. Front. Cardiovasc. Med. 5: 154.

Szarka, A., Rigó, J., Lázár, L., Beko, G., and Molvarec, A. (2010). Circulating cytokines, chemokines and adhesion molecules in normal pregnancy and preeclampsia determined by multiplex suspension array. BMC Immunol. 11: 59.

Tada, M., Takeuchi, A., Hashizume, M., Kitamura, K., and Kano, M. (2014). A highly sensitive fluorescent indicator dye for calcium imaging of neural activity *in vitro* and *in vivo*. Eur. J. Neurosci. 39: 1720–1728.

Tam, Tam, Lamarca, B., Arany, M., Cockrell, K., Fournier, L., Murphy, S., Jr., M, J.N., Granger, J.P., and Granger, J.P. (2011). Role of reactive oxygen species during hypertension in response to chronic anti- angiogenic factor (Sflt-1) excess in pregnant rats. Am. J. Hypertens. 24: 110–113.

Tanaka, M., Borgeld, H., Zhang, J., Muramatsu, S., Gong, J., Yoneda, M., Maruyama, W., Naoi, M., and Yagi, K. (2002). Gene therapy for mitochondrial disease by delivering restriction endonuclease SmaI into mitochondria. J. Biomed. Sci. 9, https://doi.org/10.1159/000064726.

Taylor, R.C. and Dillin, A. (2011). Aging as an event of proteostasis collapse. Cold Spring Harb. Perspect. Biol. 3: a004440.

Tenório, M.B., Ferreira, R.C., Moura, F.A., Bueno, N.B., Goulart, M.O.F., and Oliveira, A.C.M. (2018). Oral antioxidant therapy for prevention and treatment of preeclampsia: meta-analysis of randomized controlled trials. Nutr. Metabol. Cardiovasc. Dis. 28: 865–876.

Thadhani, R., Hagmann, H., Schaarschmidt, W., Roth, B., Cingoez, T., Karumanchi, S.A., Wenger, J., Lucchesi, K.J., and Benzing, T. (2016). Removal of soluble fms-like tyrosine kinase-1 by dextran sulfate apheresis in preeclampsia. J. Am. Soc. Nephrol. 27: 903–913.

Thadhani, R., Kisner, T., Hagmann, H., Bossung, V., Noack, S., Schaarschmidt, W., Jank, A., Kribs, A., and Benzing, T. (2011). Pilot study of extracorporeal removal of soluble Fms-like tyrosine kinase 1 in preeclampsia. Circulation 124: 940–950.

The genetics of pre-eclampsia and other hypertensive disorders of pregnancy (2011). Best Pract. Res. Clin. Obstet. Gynaecol. 25: 405–417.

Thilaganathan, B. and Kalafat, E. (2019). Cardiovascular system in preeclampsia and beyond. Hypertension 73: 522–531.

Tischner, C. and Wenz, T. (2015). Keep the fire burning: current avenues in the quest of treating mitochondrial disorders. Mitochondrion 24: 32–49.

Toescu, V., Nuttall, S.L., Martin, U., Kendall, M.J., and Dunne, F. (2002). Oxidative stress and normal pregnancy. Clin. Endocrinol. 57: 609–613.

Torbergsen, T., øian, P., Mathiesen, E., and Borud, O. (1989). Pre-Eclampsia-A mitochondrial disease? Acta Obstet. Gynecol. Scand. 68: 145–148.

Touré, V., Le Novère, N., Waltemath, D., and Wolkenhauer, O. (2018). Quick tips for creating effective and impactful biological pathways using the Systems Biology Graphical Notation. PLoS Comput. Biol. 14: e1005740.

Townley-Tilson, W.H.D., Wu, Y., Ferguson, J.E., and Patterson, C. (2014). The ubiquitin ligase ASB4 promotes trophoblast

differentiation through the degradation of ID2. PloS One 9, https://doi.org/10.1371/journal.pone.0089451.

Tranquilli, A.L., Dekker, G., Magee, L., Roberts, J., Sibai, B.M., Steyn, W., Zeeman, G.G., and Brown, M.A. (2014). The classification, diagnosis and management of the hypertensive disorders of pregnancy: a revised statement from the ISSHP. Pregnancy Hypertens. 4: 97–104.

Trogstad, L., Magnus, P., and Stoltenberg, C. (2011). Pre-eclampsia: risk factors and causal models. Best Pract. Res. Clin. Obstet. Gynaecol. 25: 329–342.

Varga, Z., Flammer, A.J., Steiger, P., Haberecker, M., Andermatt, R., Zinkernagel, A.S., Mehra, M.R., Schuepbach, R.A., and Moch, H. (2020). Endothelial cell infection and endotheliitis in COVID-19. Lancet 395: 1417–1418.

Vasileiou, P.V.S., Evangelou, K., Vlasis, K., Fildisis, G., Panayiotidis, M.I., Chronopoulos, E., Passias, P.-G., Kouloukoussa, M., and Havaki, S. (2019). Mitochondrial homeostasis and cellular senescence. Cells 8: 1–25.

Vento-tormo, R., Efremova, M., Botting, Rachel A., Turco, M.Y., Vento-tormo, M., Meyer, K.B., Park, J., Stephenson, E., and Teichmann, S.A. (2018). Single-cell reconstruction of the early maternal-fetal interface in humans. Nature 563, https://doi.org/10.1038/s41586-018-0698-6.

Videm, V. and Albrigtsen, M. (2008). Soluble ICAM-1 and VCAM-1 as markers of endothelial activation. Scand. J. Immunol. 67: 523–531.

Vishnyakova, P.A., Volodina, M.A., Tarasova, N.V., Marey, M.V., Kan, N.E., Khodzhaeva, Z.S., Vysokikh, M.Y., and Sukhikh, G.T. (2017). Alterations in antioxidant system, mitochondrial biogenesis and autophagy in preeclamptic myometrium. BBA Clin. 8: 35–42.

Wang, Liu, Y.J., Duan, G.L., Zhao, W., Li, X.H., Zhu, X.Y., and Ni, X. (2014). CBS and CSE are critical for maintenance of mitochondrial function and glucocorticoid production in adrenal cortex. Antioxidants Redox Signal. 21: 2192–2207.

Wang and Walsh, S.W. (1998). Placental mitochondria as a source of oxidative stress in pre-eclampsia. Placenta 19: 581–586.

Wang, Zhang, S., Li, Y., Wang, R., Tong, L., Wang, Y., Liu, W., Su, D., and Li, L. (2017). LY333531, a PKCβ inhibitor, attenuates glomerular endothelial cell apoptosis in the early stage of mouse diabetic nephropathy via down-regulating swiprosin-1. Nat. Publish. Group 38: 1009–1023.

Ward, M.W., Huber, H.J., Weisová, P., Düssmann, H., Nicholls, D.G., and Prehn, J.H.M. (2007). Mitochondrial and plasma membrane potential of cultured cerebellar neurons during glutamate-induced necrosis, apoptosis, and tolerance. J. Neurosci. 27: 8238–8249.

Watanabe, Y. and Dvorak, H.F. (1997). Vascular permeability factor/vascular endothelial growth factor inhibits anchorage-disruption-induced apoptosis in microvessel endothelial cells by inducing scaffold formation. Exp. Cell Res. 233: 340–349.

Weissgerber, T.L. and Mudd, L.M. (2015). Preeclampsia and diabetes. Curr. Diabetes Rep. 15, https://doi.org/10.1007/s11892-015-0579-4.

Westerhoff, H.V., Lolkema, J.S., Otto, R., and Hellingwerf, K.J. (1982). Thermodynamics of growth non-equilibrium thermodynamics of bacterial growth the phenomenological and the Mosaic approach. BBA Rev. Bioenerg. 683: 181–220.

Wickens, D., Lunec, J., Ball, G., and Dormandy, T.L. (1981). Free-radical oxidation (peroxidation) products in plasma in normal and abnormal pregnancy. Ann. Clin. Biochem. 18: 158–162.

Williams, M.A., Sanchez, S.E., Ananth, C.V., Hevner, K., Qiu, C., and Enquobahrie, D.A. (2013). Maternal blood mitochondrial DNA copy number and placental abruption risk: results from a preliminary study. Int. J. Mol. Epidemiol. Genet. 4: 120–127.

Wong, S.F., Chow, K.M., and Swiet, M. (2003). Severe acute respiratory syndrome and pregnancy. BJOG 110: 641–642.

Woods, A.K., Hoffmann, D.S., Weydert, C.J., Butler, S.D., Zhou, Y., Sharma, R.V., and Davisson, R.L. (2011). Adenoviral delivery of VEGF 121 early in pregnancy prevents spontaneous development of preeclampsia in BPH/5 mice. Hypertension 57: 94–102.

Woods, Perez-Garcia, V., and Hemberger, M. (2018). Regulation of placental development and its impact on fetal growth—new insights from mouse models. Front. Endocrinol. 9: 570.

Woolley, J.F., Stanicka, J., and Cotter, T.G. (2013). Recent advances in reactive oxygen species measurement in biological systems. Trends Biochem. Sci. 38: 556–565.

Xu, S., Tao, H., Cao, W., Cao, L., Lin, Y., Zhao, S.M., Xu, W., Cao, J., and Zhao, J.Y. (2021). Ketogenic diets inhibit mitochondrial biogenesis and induce cardiac fibrosis. Signal Transduct. Target. Ther. 6: 1–13.

Yaling, Z., Youxia, L., Yuanyuan, Q., Xiaoqing, L., Jingge, G., Xingchen, Y., Yazhuo, C., Xinnian, W., Shan, L., and Zhanzheng, Z. (2020). The soluble VEGF receptor sFlt-1 contributes to endothelial dysfunction in IgA nephropathy. PloS One 15.

Yazdi, F.T., Clee, S.M., and Meyre, D. (2015). Obesity genetics in mouse and human: back and forth, and back again. Peer J. 2015, https://doi.org/10.7717/peerj.856.

Zhai, Y., Liu, Y., Qi, Y., Long, X., Gao, J., Yao, X., Chen, Y., Wang, X., and Zhao, Z. (2020). The soluble VEGF receptor sFlt-1 contributes to endothelial dysfunction in IgA nephropathy. PloS One 15: e0234492.

Zhang, D., Wang, Tian, X., Zhang, L., Yang, G., Tao, Y., Liang, C., Li, K., and Jin, H. (2018). The increased endogenous sulfur dioxide acts as a compensatory mechanism for the downregulated endogenous hydrogen sulfide pathway in the endothelial cell inflammation. Front. Immunol. 9: 882.

Zhao, R.Z., Jiang, S., Zhang, L., and Yu, Z. Bin (2019). Mitochondrial electron transport chain, ROS generation and uncoupling (Review). Int. J. Mol. Med. 44: 3–15.

Zhong, W., Gao, X., Wang, S., Han, K., Ema, M., Adams, S., Adams, R.H., Rosenblatt, M.I., and Azar, D.T. (2017). Prox1-GFP/Flt1-DsRed transgenic mice: an animal model for simultaneous live imaging of angiogenesis and lymphangiogenesis. Angiogenesis 20: 581–598.

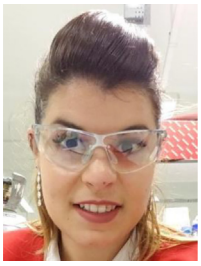
Zhu, C., Yu, Y., Montani, J.P., Ming, X.F., and Yang, Z. (2017). Arginase-I enhances vascular endothelial inflammation and senescence through eNOS-uncoupling. BMC Res. Notes 10: 1–8.

Zhu, J., Lim Jethro, J.L., Li, B., Mergenthal, A., Rayes, A.A., Tang, H., Berger, T.W., and Bouteiller, J.M.C. (2020). A computational model of mitochondria motility in axons. Proceedings of the annual international conference of the IEEE engineering in medicine and biology society. EMBS, pp. 2287–2290, https://doi.org/10.1109/EMBC44109.2020.9176609.

Zorov, D.B., Juhaszova, M., and Sollott, S.J. (2014). Mitochondrial reactive oxygen species (ROS) and ROS-induced ROS release. Physiol. Rev. 94: 909–950.

**Supplementary Material:** The online version of this article offers supplementary material (https://doi.org/10.1515/hsz-2021-0121).

## Bionotes



**Yolanda Correia**
Aston Medical School, College of Health & Life Sciences, Aston University, Aston Triangle, Birmingham B4 7ET, UK
**y.correia@aston.ac.uk**

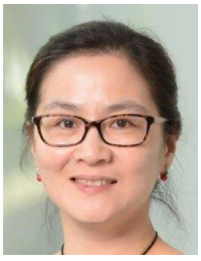
Yolanda Correia is a PhD student at Aston Medical School, Aston University. She previously studied Forensic Science at Kingston University and got her Masters at Virginia Commonwealth University. Currently, her research focuses on using murine models to seek a deeper understanding of the pathophysiology of preeclampsia.

**Julia Scheel**
Department of Systems Biology and Bioinformatics, University of Rostock, D-18051 Rostock, Germany
**julia.scheel@uni-rostock.de**
**https://orcid.org/0000-0002-0034-7755**

Julia Scheel is a PhD student at the department of Systems Biology and Bioinformatics, Rostock University. She studied Biology at the University Erlangen-Nuremberg and received a MSc in Immunology at University of Aberdeen and a MSc in Neurobiology at University of Tübingen. Her current research focuses on applying systems biology approaches to identify biomarkers and candidates for therapeutic targets for placental diseases.

**Shailendra Gupta**
Department of Systems Biology and Bioinformatics, University of Rostock, D-18051 Rostock, Germany

Shailendra Gupta is a scientist and group leader at Department of Systems Biology and Bioinformatics, University of Rostock, Germany. His methodologies range from the construction and analysis of large-scale disease maps to structural and molecular level investigation of regulatory biomolecules. His tools are used for therapy personalisation, target prediction, drug discovery and drug repositioning. More information on his research projects and methodologies can be found at www.sbi.uni-rostock.de/team/detail/shailendra-gupta.



**Keqing Wang**
Aston Medical School, College of Health & Life Sciences, Aston University, Aston Triangle, Birmingham B4 7ET, UK

Keqing Wang is a Lecturer at Aston Medical School, Aston University. She has extensive research experience in immunology, inflammation and the vascular biology of pregnancy. Her research focuses on better understanding of role of gaseous transmitters in the pathogenesis of cardiovascular diseases related to pregnancy. The ongoing work is aimed at the development of novel therapies and diagnostic tools for these disorders.