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Chapter 6

Soluble fms-like tyrosine kinase 1 and soluble endoglin are elevated circulating anti-angiogenic factors in pre-eclampsia

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Abstract

Pre-eclampsia, characterized by hypertension and proteinuria, affects approximately 3–5% of all pregnancies worldwide and is a major cause of maternal and fetal morbidity and mortality. Maternal endothelial dysfunction is associated with disease pathogenesis. Recently, reports have shown that elevated levels of circulating soluble fms-like tyrosine kinase 1 [sFlt1] and soluble endoglin [sEng] are associated with pre-eclampsia. Flt1 is a receptor for vascular endothelial growth factor receptor [VEGF], whereas endoglin [Eng] is an auxiliary receptor for transforming growth factor-β [TGF-β] super-family members. Both signalling pathways modulate angiogenesis and are involved in vascular homeostasis. Increased levels of sFlt1 and sEng dysregulate VEGF and TGF-β signalling respectively, resulting in endothelial dysfunction of maternal blood vessels. This review summarizes our current knowledge of Flt1 and endoglin and soluble forms in pre-eclampsia. Furthermore, it highlights the predictive and early-screening value of circulating levels of sFlt1 and sEng for the risk of developing pre-eclampsia

Keywords: Angiogenesis, Endothelial dysfunction, Pre-eclampsia, sFlt1, Soluble endoglin, TGF-β, VEGF

Introduction

Pre-eclampsia is a pregnancy-specific syndrome that develops after mid-gestation and is characterized by de novo hypertension [systolic blood pressure P140 mm Hg or diastolic blood pressure P90 mm Hg] accompanied by new onset proteinuria defined as 300 mg of protein/24 h or more after 20 weeks of gestation [1]. To date, the clinical incidence of pre-eclampsia is 3–5% of pregnancies, which in some instances can lead to complications such as maternal renal failure, hemolysis, elevated liver enzymes and low platelets [HELLP], seizures, liver failure, stroke or death [2–4]. For the fetus, pre-eclampsia can result in intrauterine growth restriction [IUGR], preterm delivery and death [2–4]. Pre-eclampsia resolves after placental delivery; however, women with a history of pre-eclampsia are at higher risk of developing cardiovascular complications later in life [5,6].

The etiology of pre-eclampsia remains incompletely understood. Its origin lies in defective placentation resulting in the release of placenta-derived angiogenic modulators into the maternal circulation. Studies have shown that levels of soluble fms-like tyrosine kinase 1 (sFlt1) and soluble endoglin (sEng) are elevated in the blood of pregnant women suffering from pre-eclampsia [7–9]. Flt1 is a vascular endothelial growth factor [VEGF] receptor and also known as VEGF receptor 1 (VEGFR1), while endoglin is a co-receptor for transforming growth factor (TGF)-β super-family members. Both receptors are highly expressed on endothelial cells and are involved in maintaining vascular homeostasis. The aberrantly large quantities of these placenta-derived soluble receptors in pre-eclamptic women can cause maternal endothelial cell dysfunction and thereby the resulting clinical symptoms of pre-eclampsia.

Besides the contribution of these anti-angiogenic factors, inflammatory responses and metabolic syndromes are also considered to be associated with the etiology of pre-eclampsia [1,10]. In this review we focus on discussing the roles of placenta-derived sFlt1 and sEng in endothelial dysfunction and their potential as biomarkers to assess the risk of developing pre-eclampsia.

The biology of sFlt1 and sEng

The biology of sFlt1

VEGF is a multifunctional molecule that is involved in numerous biological processes during postnatal development and in both physiological and pathological conditions. The mammalian VEGF family is composed of VEGF- A, VEGF-B, VEGF-C VEGF-D and placenta growth factor (PlGF). The predominant form, VEGF-A, is alternatively spliced in different isoforms which have pro- and anti- angiogenic properties [11]. Three types of VEGF receptors are activated upon ligand binding,

namely Flt1, Flk1 (VEG- FR2) and Flt4 (VEGFR3). Co-receptors, such as neuropilins and heparan sulfate proteoglycans modify VEGFR activities in diverse fashions [12–16]. Flt1 binds VEGF-A, -B and PlGF, whereas Flk1 binds to VEGF-A and -C and Flt4 binds VEGF-C and -D [17].

VEGFs and their receptors are crucial for vascular development and lymphatic vessel formation. VEGFRs consist of an extracellular domain, a trans-membrane domain and an intracellular protein tyrosine kinase domain. Ligand binding leads to VEGFR dimerization and activation of the tyrosine kinase activity, upon which diverse intracellular signalling pathways are initiated that modulate vascular biological functions. Flt1 and Flk1 mediated signalling events regulate endothelial cell function, such as endothelial cell proliferation, migration, tube formation and branching [18]. VEGFR3 signalling is mainly important for lymphatic vessel development [19].

The importance of Flk1 and Flt1 in vascular function became apparent from the analysis of knockout mice. Depletion of either Flt1 or Flk1 in mice resulted in embryonic lethality due to vascular malfunction, but with distinct differences in the phenotypes. Flt1 knockout mice showed irregular vascular organization due to endothelial cell overgrowth, whereas Flk1 knockout embryos had an absence of blood vessels and blood islands [20,21]. The extracellular domain of both Flk1 and Flt1 consists of seven extracellular immunoglobulin (Ig) homology domains. The 2nd Ig homology domain is sufficient for VEGF binding to Flt1, while Flk1 requires both the 2nd and 3rd Ig homology domains for adequate VEGF binding [22–25]. VEGF-A has a higher affinity for Flt1 than Flk1, which may be explained by the difference in ligand-binding domains. Yet the kinase activity of Flt1 in response to VEGF-A binding is weak [26–28], possibly because the juxtamembrane domain of the intracellular domain represses kinase activation [29]. Interestingly, mice deficient in the kinase domain of membrane-anchored Flt1, but with an intact ligand binding domain, revealed no defects in vascular development, suggesting the kinase activity of Flt1 is dispensable for its role in vasculature formation [30]. Taken together, these data suggest that Flt1 may function as a decoy receptor for VEGF-A, thereby regulating VEGF-A bio-availability for Flk1 [18]. PlGF binds directly to Flt1 and amplifies VEGF-driven angiogenesis through Flk1 [31]. PlGF can outcompete VEGF-A for the binding of Flt1, indirectly promoting VEGF-A/Flk1 signalling by increasing the bio-availability of VEGF-A for Flk1 [32,33]. Nonetheless, other experimental evidence indicates that Flt1 not only serves to fine-tune the VEGF-Flk1 axis, but is also directly involved in signal transduction. VEGF-A and PlGF induce Flt1 receptor phosphorylation and downstream signalling [31]. Loss of Flt1 kinase activity shows deficient inflammation and angiogenesis in various disease models such as atherosclerosis, choroidal neovascularization, lung metastasis and rheumatoid arthritis [34–37]. In addition, Flt1 can act as a negative regulator for endothelial tip cell sprouting and branching morphogenesis in zebra fish system [38]. Up to date, the apparent function of Flt1 in angiogenesis is evident, but the exact signalling of Flt1 remains to be elucidated.

The transmembrane Flt1 protein is encoded by 30 exons; sFlt1 is generated through alternative splicing (Fig. 1A) and consists of exons 1 through 13 with an intron 13 derived encoded carboxy tail [39–41]. sFlt1 and Flt1 share the first six Ig-homology domains, which are essential for ligand binding [42,43]. The splicing of sFlt1 is possibly regulated by hypoxia and histone jumonji domain-containing demethylases [44–46]. Recent studies have described additional splice variants of sFlt1, (termed as sFlt1-v2,-v3,-v4). While all share an extracellular domain followed by a short C-terminal tail, they differ in composition [47,48]. Different from the other variants, sFlt1-v2 has been reported to be highly expressed in placental tissues as a non-endothelial cell form of sFlt1 [48,49]. However the relevance of this secreted sFlt1 on placenta function *in vivo* has not yet been investigated. In addition to alternative splicing, a ligand-induced proteolytically cleaved version of soluble Flt1 has been found to be expressed by leukemic cells [50]. Whether proteolytic shedding also contributes to sFlt1 in pre-eclampsia is not known.

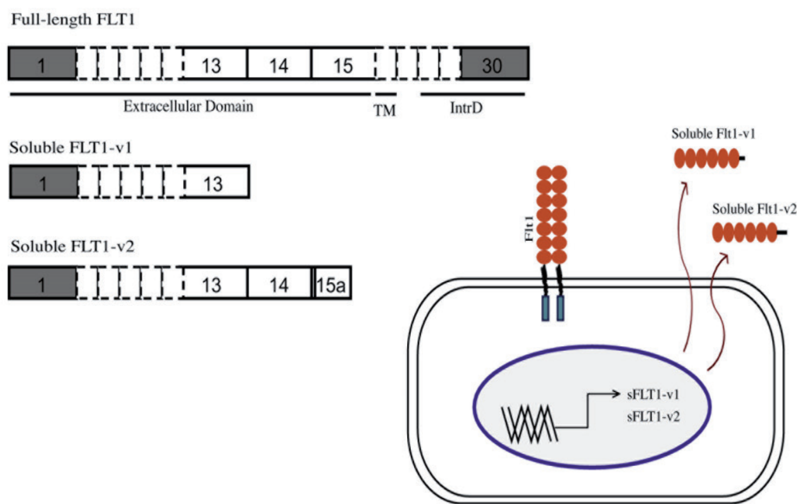

Fig.1A. *Different transcripts of sFlt1 and the generation of sFlt. The transmembrane VEGF receptor, including the extracellular domain, transmembrane domain (TM), and intracellular catalytic domain (IntrD), is encoded by all 30 exons of the FLT1 gene. The sFLT1 proteins are the result of alternative splicing, generating a secreted form of FLT1 with a unique 31 amino acid (sFLT1-v1) or 28 amino acid (sFLT1-v2) c-terminal tail. In human placenta, 80% of FLT1 expression corresponds to transcript sFLT1-v2, 15% to sFLT1-v1, and 5% to the transmembrane version.*

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sFlt1 sequesters circulating VEGF-A, VEGF-B and PlGF [31,42] (Fig.1B). In addition, it can form a stable receptor complex with the extracellular domain of Flk1, thereby interfering with Flk1 dimerization and subsequent intracellular signalling [43]. Due to the neutralizing function of sFlt1 for its natural ligands, sFlt1-based trap has been developed for preclinical purpose, which contains the first three Ig-homology domains of Flt1 [51]. sFlt1 and the sFlt-based trap have been shown to act as negative regulators for endothelial function [52] and angiogenesis in several pre-clinical studies [53–56]. Notably, Flt1 and sFlt1 are highly expressed in placental tissues, particularly in the trophoblast [49,57–59] and can be released into the maternal circulation [60].

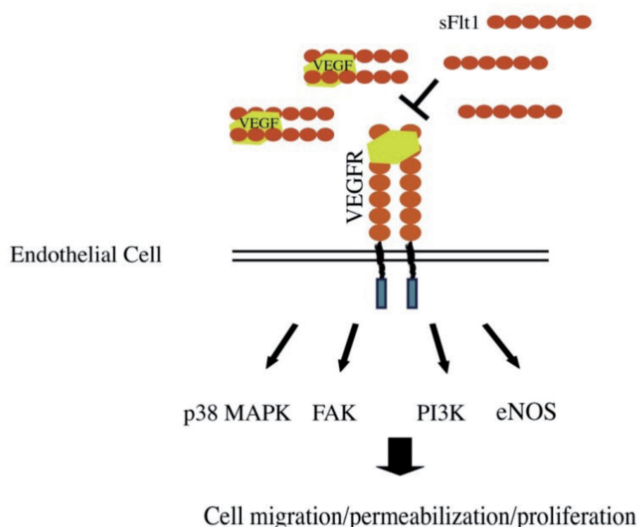

Fig.1B. *VEGF/Flt1 signalling and effect of sFlt1 thereon. VEGF induces Flt1 receptor dimerization. As a result of Flt1 receptor activation, downstream signalling pathways are indirectly or directly activated, such as p38 mitogen-activated protein kinases (p38 MAPK), focal adhesion kinase (FAK), phosphoinositide 3-kinase (PI3K), endothelial nitric oxide synthase (eNOS). The VEGF/Flt1 system regulates endothelial cell migration, proliferation and permeabilization. Increased sFlt1 levels in the circulation can sequester VEGF locally and thereby interfere with VEGF-induced endothelial function.*

The biology of sEng

Endoglin (CD105) is a membrane-bound glycoprotein which functions as an auxiliary/co-receptor for TGF-β super-family members [61]. The TGF-β superfamily includes TGF-βs, activins and bone morphogenetic proteins (BMPs); they affect not only vascular cells, but also many other cell types. The TGF-β superfamily signals via complexes of type I and type II serine/threonine kinase receptors [62].

Endoglin contains an extracellular domain, a single transmembrane domain and a short cytoplasmic domain [63] (Fig. 2A). The extracellular domain of endoglin harbors a signal peptide, an orphan domain and a zona pellucida (ZP) domain. The ZP domain is involved in endoglin oligomerization; the extracellular and intracellular domains mediate heteromeric interactions with TGF-β receptors I and II (TβRI/II) [64]. TGF-β only binds to endoglin in complex with TβRII, whereas BMP-9 directly binds to the orphan domain of endoglin [65,66]. Endoglin is closely related to the TGF-β co-receptor betaglycan. Endoglin binds to TGF-β1 and β3, while betaglycan interacts with all three TGF-β isoforms [67]. In part, the cell type-specific effects of TGF-β are mediated by the auxiliary receptors that demonstrate differential expression patterns and interact with different affinity to TGF-β isoforms. Due to differential mRNA splicing, two membrane-bound forms of endoglin have been described: long-form endoglin (L-endoglin) and short-form endoglin (S-endoglin). L-endoglin is the most abundantly expressed isoform [68,69]. Apart from the membrane-bound forms, endoglin can exist as a soluble form (sEng) (Fig. 2A).

Endoglin is expressed at low levels in quiescent endothelial cells, but is highly expressed in proliferating endothelial cells [70]. In addition to endothelial cells, endoglin is also expressed in syncytiotrophoblasts [70,71], stromal cells [71,72] and hematopoietic cells [73] of full term placentas. Endoglin expression can be induced and regulated by hypoxia, TGF-β1, TGF-β3 and BMP-9 [66,75,76]. Mouse aortic endothelial cells (MAEC) isolated from endoglin null mice display reduced proliferation, migration, VEGF secretion and decreased endothelial nitric oxide synthase (eNOS) expression [77–79]. Lebrin *et al.* also found that endothelial cells derived from endoglin null embryos demonstrated impaired proliferation in culture [61]. Up-regulation of endoglin has been shown to protect endothelial cells from TGF-β1-induced apoptosis [79]. The importance of endoglin in vascular biology was demonstrated by the embryonic lethality of mice deficient in endoglin due to defective angiogenesis *in vivo* [80,81]. Mutations in endoglin cause the autosomal dominant disease hereditary hemorrhagic telangiectasia [HHT] type I. HHT is a vascular dysplasia disease characterized by the development of mucocutaneous telangiectasias and arteriovenous malformations in the brain, lungs, liver and gastrointestinal tract. Endoglin heterozygous mice exhibit vascular lesions due to capillary malformation, resembling HHT type 1 [81–83]. Thus, endoglin has a pivotal role in endothelial cell function [74,84].

Levels of sEng are elevated in the sera of pre-eclamptic patients [7,9] as well as in colorectal and breast cancer patients [85,86], which results in abnormal angiogenic responses. It has been reported that sEng is generated via metalloproteinase (MMP)-14 (MT1-MMP) mediated shed- ding of membrane bound endoglin in colorectal cancer, at the site close to the transmembrane domain of endoglin [86] (Fig.2A). This cleaved sEng contains the entire extracellular domain, retaining the ability of binding to TGF-β and BMP-9 [9,66]. Thus, local shedding of endoglin is a potential source of sEng, which subsequently can affect tumor angiogenesis in the tumor microenvironment. MMP14 was recently shown to be involved in the generation of sEng in pre-eclampsia patients. MMP14 was expressed by syncytiotrophoblasts and interacted with endoglin within the pre-eclamptic placenta. Challenging trophoblasts with MMP14 inhibitors attenuated the production of sEng [87].

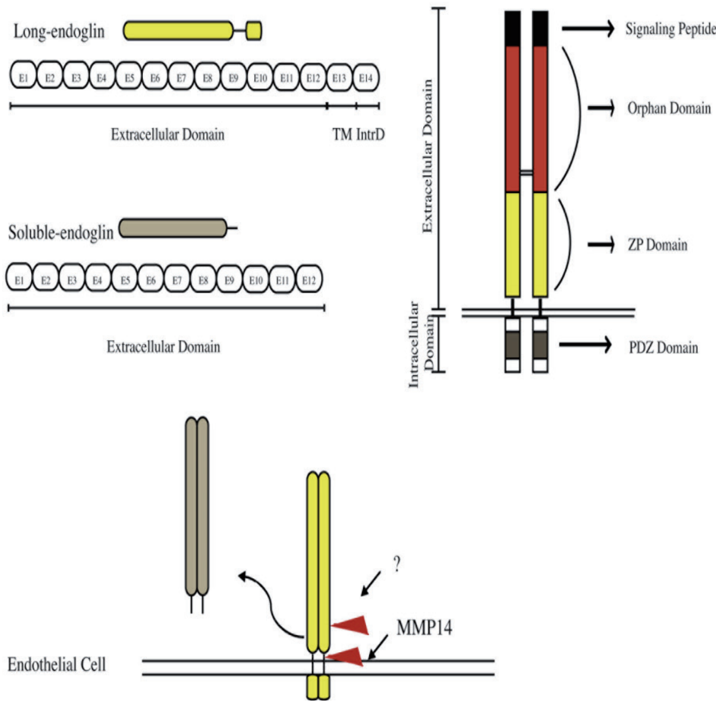

Fig. 2A. *Different forms of endoglin and the generation of sEng. Endoglin is a 90kDa protein that contains a large extracellular domain and short intracellular domain that lacks an enzymatic motif. Endoglin is a dimeric protein and monomers are connected by a disulphide bridge. Several sub-domains have been identified, including orphan domain and TGF-β/BMP binding domains, receptor interacting domains. The long form (L; yellow) and the soluble form (S; gray) forms differ in the size of their intracellular domain. At its carboxy terminus endoglin has a PDZ interaction motif. sEng consists of part of the extracellular domain of endoglin; it lacks a transmembrane and intracellular domain. sEng can be generated by shedding of membrane bound endoglin possibly via MMP14 activity. It is not excluded that endoglin can be cleaved by any other unknown enzymes because it contains multiple potential proteolytic cleavage site.*

It has been shown that sEng inhibits angiogenesis in ex vivo assays, such as the fetal mouse metatarsal [86] and chick chorioallantoic membrane assay [88]. sEng can exert anti-angiogenic effects on endothelial cells by modulating TGF-β/BMP signalling [9,86,88,89] (Fig. 2B); Studies *in vitro* have shown that sEng interferes with TGF-β signalling and eNOS activity [9]. The explanation for these findings could be that sEng functions as a scavenger for circulating ligands such as TGF-β1/3 and BMP-9 [88], and thereby affects the vascular balance maintained by TGF-β/BMP signalling. It is also possible that by binding to TGF-β receptors, sEng interferes with the availability of TGF-β receptors for membrane-bound endoglin [88].

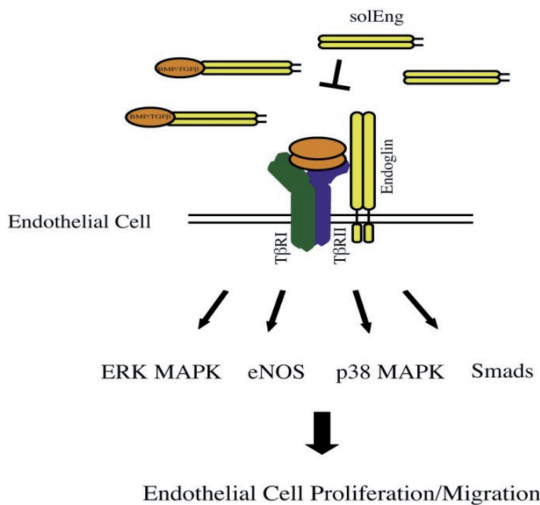

Fig.2B. *TGF-β/BMP/endoglin signalling and the effect of sEng thereon. TGF-β and BMP bind to heteromeric type II/type I serine/threonine kinase receptor complexes. Endoglin is an auxillary receptor and can interact with, and modulate ligand binding to type II/type I receptor complexes. The active receptor complex can activate intracellular Smad pathways and non-Smad pathways: early response kinase (ERK); phosphoinositide 3-kinase (PI3K); p38 mitogen-activated protein kinases (p38 MAPK), endothelial nitric oxide synthase (eNOS). In the endothelial cell, TGF-β/ BMP signalling modulates cell migration and proliferation. sEng functions as a negative regulator for TGF-β/BMP signalling in endothelial cells.*

Endothelial cell dysfunction with increased levels of sFlt1 and sEng in pre-eclampsia

Angiogenesis is tightly controlled by pro- and anti-angiogenic factors. An imbalance in this process can lead to excessive or insufficient angiogenic responses, which has been associated with different diseases. Studies in humans and animal models have demonstrated that TGF-β and VEGF signalling play crucial roles in maintaining physiological vascular homeostasis by modulating endothelial cell function [90–92].

Initial insights from human studies

In 2003, the first studies were published that clearly demonstrated increased sFlt1 levels in the circulation of pre-eclamptic women as to compared normotensive pregnant women [8,92]. Also, free VEGF and PlGF levels were lower in the blood of pre-eclamptic patients compared to

normotensive pregnancies [8,93], confirming the antagonistic effects of sFlt1 on the VEGF system. Subsequently, many other studies have confirmed these findings with the agreement that sFlt1 holds predictive value reviewed by Lapaire [94]. Most importantly, the increase in sFlt1 was detected before the onset of pre-eclampsia. A human study performed by Levine and colleagues has shown the significant predictive value for sFlt1 [7]. Sera were analyzed for sFlt1 by ELISA from a total number of 120 women with pre-eclampsia and matched healthy pregnant women during their pregnancy. Women who developed pre-eclampsia showed higher levels of sFlt1 compared with the control group. An increase in sFlt level was detected 5 weeks before the clinical onset of pre-eclampsia. Meanwhile, PlGF concentration was significantly lower in women who later developed pre-eclampsia compared to controls [95]. Because the increase of sFlt1 is accompanied with a decrease in PlGF levels, the angiogenic balance was most likely shifted toward inhibition in women with a high risk of developing pre-eclampsia. This subverted anti-angiogenic status in pre-eclampsia patients is further supported by a renal biopsy study, which illustrated mild glomerular endotheliosis in normal pregnancy, but severe lesions in pre-eclampsia [96].

A landmark study, performed by Venkatesha et al., showed that the expression of sEng is elevated both in placenta tissues as well as serum of pre-eclamptic patient compared to controls. The high levels of sEng in pre-eclamptic patients were dramatically reduced after placental delivery, indicating that the increase in sEng is pregnancy-associated. Similar to the role of excessive sFlt1 in kidney dysfunction, elevated sEng levels induce glomerular endotheliosis [9,97].

Insights from rodent studies

Many of the clinical symptoms of pre-elampsia can be recapitulated in animal models. As the mouse placenta shares molecular and structural similarities with the human placenta reviewed by Rossant [98], a variety of rodent models have been proposed for pre-eclampsia studies. These models include the sFlt1/sEng induction model, the reduced uterine perfusion pressure (RUPP) model and the renin angiotensin abnormalities (AT-AAs) model.

Rats administered with adenoviral sFlt1 showed a clear increase in blood pressure and proteinuria recapitulating major features observed in human patients [8]. Similar responses were observed after intravenous injection of sFlt1 protein into mice [99]. Introduction of adenoviral sEng into pregnant rats also resulted in a significant increase in mean arterial pressure, mild renal endotheliosis and vascular damage in the placenta [9,100]. Furthermore, the combination of adenoviral induction of sEng and sFlt1 in pregnant rats increased complications of pre-eclampsia, including infarction at the maternal-fetal interface, fetal growth restriction, hepatic ischemia and necrosis [9]. Reversely, neutralization of free sFlt1 in pre-eclamptic mice by the increase of VEGF or PlGF ameliorated the pre-eclampsia syndrome [99,101].

In the RUPP model, the bilateral utero-ovarian arteries are ligated in pregnant animals. This results in the development of pre-eclampsia-like symptoms such as hypertension, proteinuria and glomerular endotheliosis [102]. This technique has been applied in different animals including rabbits [103], dogs [104,105], monkeys [106], baboons [107] and sheep [109] to study hypertension in relation to blood pressure changes during pregnancy. In a recent study, using the RUPP model to induce pre-eclamptic hypertension in rats, it was shown that both the sFlt1 and the sEng levels [99,108] in serum were significantly increased compared to the control group, again confirming the association of elevated sFlt1/sEng levels with pre-eclamptic hypertension.

Another mouse model to induce pre-eclampsia is achieved by the injection of angiotensin receptor activating autoantibodies (AT1-AAs), which are obtained from women with pre-eclampsia, into healthy pregnant mice. When mice were co-treated with an AT1 receptor antagonist [losartan], pre-eclampsia did not develop [9,110] validating the importance of AT1-AAs for pre-eclampsia onset. The injection of AT1-AAs resulted in elevated levels of sFlt1 and sEng in pregnant mice, whereas these levels were not affected in non-pregnant AT1-AA treated control mice [110,111], further suggesting the relevance of sFlt and sEng in pre-eclampsia. Similar to human studies, increased levels of sFlt1 and sEng are associated with pre-eclampsia in rodent models. These models provide researchers with great opportunities to investigate the intervention value of sFlt1 and sEng as potential therapeutic targets.

sFlt1 and sEng as circulating biomarkers for pre- eclampsia

As hypertension and proteinuria are not specific symptoms for pre-eclampsia, a specific diagnostic marker is greatly needed for pre-eclamptic patients for early diagnosis and prevention. For this purpose a number of biomarkers have been under investigation [112]. The expression of sFlt1 and sEng is increased in pre-eclamptic patients prior to the occurrence of the pre-eclamptic syndrome, indicating their predictive value for the onset of pre-eclampsia. Nevertheless, the level of sFlt1 during pregnancy is variable between individuals, so it is difficult to interpret its accuracy as a circulating biomarker. Therefore, recent studies have analyzed the sFlt1/PlGF ratio to enhance the sensitivity of this marker as an indicator for the onset of pre-eclampsia [7]. This ratio increases before the onset of pre-eclampsia, but it does not distinguish between women with subsequent gestational hypertension and the control group [7]. Using an automated sFlt1/PlGF assay [Elecsys® sFlt1 and PlGF assay developed by Roche] the sFlt1/PlGF ratio was shown to be a valuable tool in the assessment of pre-elampsia [112,113].

Besides increased sFlt1 levels, sEng levels are also elevated in pre-eclamptic patients with an increased sFlt1/ PlGF ratio [7]. Increased sEng has been shown to be associated with pre-eclampsia development in a cohort study [114] and supported by another independent study in a Korean cohort [115]. In addition, sEng is reported to be informative to discriminate pre-eclampsia from gestational hypertension and chronic hypertension [116]. Of note, its increase also occurred in normotensive pregnancies with IUGR [117]. However, the association between IUGR and increased sEng levels was challenged by other studies in which no association between IUGR with sEng levels could be demonstrated [118].

The currently available enzyme-linked immunosorbent assay (ELISA) kits for the detection of sFlt1 are comprised of antibodies that do not distinguish the different isoforms of sFlt1. sFlt1 variants are expressed at different levels in the placenta [49]. Use of antibodies which discriminate between the different subtypes could more specifically evaluate the potential roles of different isoforms in the pathogenesis of pre-eclampsia. With regard to sEng detection, different molecular weights of sEng have been reported. A study by Hawinkels *et al.* reported sEng to be around 80 kDa in endothelial cells, whereas Venkatesha *et al.* detected a 65 kDa form of sEng in placental tissue. The size of sEng appears to be variable in different experimental settings. To validate the prediction value of sEng, more studies are needed to understand the source of different sEng forms.

Concluding remarks and future perspectives

Pre-eclampsia is a complex pregnancy-related syndrome. Deficient placental function has been recognized as the major cause, but the pathological mechanism that underlies this disease remains less understood. Concerning the high incidence among pregnant women globally, an effective diagnostic tool for pre-eclampsia is urgently needed. Notably, the predictive potential of sFlt1 and sEng has drawn great attention in the research field of pre-eclampsia in recent years, as substantial evidence implies that aberrant sFlt1 and sEng levels are involved in the angiogenic imbalance in pre-eclampsia patients. Whereas the application of combining sFlt1/PlGF ratio together with the sEng concentration as diagnostic markers may contribute to better prediction and early identification of pre-eclamptic patients, further studies regarding sFlt1 and sEng as predictive biomarkers for pre-eclampsia are required. Furthermore, it would not be surprising if aberrant sEng or sFlt1 levels are associated with other diseases, such as vascular complications in hypertension, diabetes and gestational proteinuria patients [119,120].

To explore potential therapeutic approaches for suppressing of pre-eclampsia progression the perturbed VEGF-sFlt1 balance was restored in mice by the administration of VEGF. This indeed alleviated the pre-eclampsia-like symptoms [101]. Targeting endoglin shedding may be an option to suppress sEng levels in the circulation. Alternatively, pharmalogically targeting of

the signalling upstream of sFlt1 and sEng expression might be more effective. Increasing levels of Heme-oxygenase-1 [HO-1] may be beneficial for ameliorating pre-eclampsia HO-1 is a negative regulator for both sFlt1 and sEng expression in human placenta explants at the transcriptional level [121,122]. This strategy has been pursued n the ongoing trial StAmP [Statin to Ameliorate Early Onset Pre-eclampsia] to validate the use of statins in early-onset pre- eclampsia [123]; statins, a class of anti-oxidant agents, induce HO-1 expression [121,124]. However, an issue which should not be neglected is that all aforementioned angiogenic-based therapeutic approaches may affect the systemic endothelial homeostasis.

Recent findings have provided evidence suggesting that levels of sFlt1 and sEng associate with pathology of pre-eclampsia. Yet understanding the exact function of sFlt1 and sEng in pre-eclampsia remains incomplete. Future advances in understanding the biology of sFlt1 and sEng in vascular biology could possibly reveal the etiology of pre-eclampsia and potentially be beneficial in diagnosing patients at risk for the development of pre-eclampsia.

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