New Aspects in the Pathophysiology of Preeclampsia

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Abstract. Preeclampsia, the de novo occurrence of hypertension and proteinuria after the 20th week of gestation, continues to exert an inordinate toll on mothers and children alike. Recent clinical trials, new physiologic insights, and novel observations on pathogenesis have altered the thinking about preeclampsia. The mechanisms surrounding relaxin and its effects on the circulation and on matrix metalloproteinases have been elucidated. The growth factor’s receptor, fms-like tyrosine kinase 1, has been shown to exist in a soluble form that is able to inactivate vascular endothelial-derived growth factor and human placental growth factor. Compelling evidence has been brought forth suggesting that fms-like tyrosine kinase 1 is a circulating factor that can cause preeclampsia. Preeclamptic women have high circulating levels of asymmetric dimethyl arginine that could account for the generalized endothelial dysfunction observed in preeclampsia. Preeclamptic women also produce novel autoantibodies that may serve to activate angiotensin receptors. These new observations raise the possibility that the treatment of preeclamptic women will soon be improved.

The term preeclampsia refers to the new onset of hypertension (>140/90 mmHg) and proteinuria after 20 wk of gestation in previously normotensive, nonproteinuric women (1). The condition is common and occurs in ~5% of pregnancies in the United States and Europe. Eclampsia is a life-threatening complication and is characterized by grand mal seizures. The term comes from the Greek word for lightning. A severe variant of preeclampsia also features hemolysis, elevated liver enzymes, and low platelets (HELLP syndrome). This condition occurs in ~1 per 1000 pregnancies. Predisposing factors are a positive family history, hypertension, diabetes, preexisting renal disease, multiple pregnancies, and a poor obstetric history. Nephrologists are often called on to see preeclamptic women because of the severe BP elevation and renal disease. Thus, new clinical or experimental information on this condition is important information for nephrologists.

Preeclampsia and Subsequent Cardiovascular Risk

The relevance of preeclampsia to the offspring is well recognized. Children who are born to preeclamptic mothers commonly have low birth weight, and their subsequent cardiovascular risk has been a vast investigational field. The mother’s outcome has attracted less interest. Chesley, the father of modern preeclampsia research, was of the opinion that once the condition was over, the mothers had no greater risk of adverse long-term outcomes than women without preeclampsia from the general population (2). This issue may prove to be the only matter in which Chesley’s opinion was erroneous. Several recent studies suggest that the converse is the case. Smith et al. (3) studied the pregnancy complications and the maternal risk of ischemic cardiac death in 129,290 births. They found that delivering an infant with low birth weight for gestational age increased the hazard ratio for ischemic heart disease or death to 1.9. Preterm delivery was associated with a risk of 1.8, compared with women who had no such delivery. The associations were additive. Preeclamptic women who delivered a small infant early, generally the rule, had a risk of hospital admission for ischemic heart disease or death seven times higher than the control women did.

A second cohort study was presented shortly thereafter. Irgens et al. (4) published the results of 626,272 live births in Norway between 1967 and 1992. They found that women who...
had had preeclampsia had a 1.2-fold higher long-term risk of all-cause death than women who did not have this condition. In women with preeclampsia and a preterm birth, the risk was 2.7-fold higher. The risk of death from cardiovascular causes was increased eightfold in preeclamptic women with a child of low birth weight. Strangely, preeclampsia seemed to protect women from death by cancer by 0.36-fold. This state of affairs resulted in the finding that women with preeclampsia had only a 1.2-fold higher long-term risk of death from any cause. Perhaps expected, the fathers did not enter into the equation. This huge cohort study provides very strong evidence that cardiovascular risk is increased in women with preeclampsia compared with control subjects, particularly when the child is born preterm and has low birth weight, both of which are generally the rule in women with preeclampsia. Wilson et al. (5) examined the relationship between preeclampsia and the risk of hypertension and stroke later in life. They selected women from a cohort who had delivered between 1951 and 1970. They found that any hypertensive disorder of pregnancy increased the later risk for hypertension and stroke. The stroke relative risk was increased in women who had been preeclamptic and was 3.59.

Women with preeclampsia develop pronounced swelling of glomerular endothelial cells and feature various deposits under these cells (6). Moreover, they display fibrin within endothelial cells and mesangial cells. The basement membrane thickens. These changes are reversible; however, the functional changes are not necessarily totally reversible. Bar et al. (7) followed a group of women who had had preeclampsia and compared these women with women who had normal spontaneous deliveries. They found that at 2 to 4 mo after delivery, two thirds of preeclamptic women had microalbuminuria. More disturbing is that 3 to 5 yr later, half of the women with the preeclampsia history still had microalbuminuria. The protein excretion at this time point was approximately fourfold higher than that of women who had not had preeclampsia. Whether the microalbuminuria persists indefinitely is uncertain. Data from Roest et al. (8) suggest that microalbuminuria in postmenopausal women is a substantial cardiovascular risk factor. We have little reason to believe that microalbuminuria in premenopausal women is any less of a risk. Microalbuminuria is a manifestation of endothelial dysfunction. Endothelial function in preeclamptic women is impaired. The impairment was still present in a recent postpartum follow-up study (9). Preeclampsia is associated with insulin resistance and elevated homocysteine levels. Evidence exists that these conditions continue after delivery and represent a long-term risk.

Sattar and Greer (10) focused on pregnancy complications and maternal cardiovascular risk and pointed out the opportunities for screening and risk reduction. Women with a history of adverse pregnancy outcome are at increased risk for cardiovascular disease later in life. Pregnancy complications and coronary heart disease may have common mechanisms. Clearly, women with gestational diabetes, a preeclampsia risk factor, must be screened for diabetes later in life. Women with low birth weight infants should be screened for cardiovascular disease in their 30s. Maternal vascular risk factors, such as higher BP, insulin resistance, obesity, excessive weight gain in pregnancy, and hyperlipidemia, correlate with increased risk of preterm delivery and low birth weight. In conclusion, asking about preeclampsia, low birth weight infants, or any adverse pregnancy outcome provides valuable information in terms of assessing cardiovascular risk in women.

Abnormal Placenta and Placental Ischemia

Preeclampsia occurs only in the presence of the placenta or a hydatidiform mole and remits dramatically postpartum after the delivery of the placenta (11). The pathogenesis of preeclampsia may involve abnormal cytotrophoblast invasion of spiral arteries, decreased uteroplacental hypoperfusion, an imbalance between increased synthesis of thromboxane and decreased production of prostaglandin I2, increased oxidative stress, disordered endothelin metabolism, or endothelial dysfunction.

During normal placental development, cytotrophoblasts invade the maternal spiral arteries and completely remodel the maternal spiral arteries into large capacitance vessels with low resistance (12). This endovascular cytotrophoblast invasion involves replacement of not only the endothelium but also the highly muscular tunica media. Furthermore, during normal differentiation, invasive trophoblasts alter their adhesion molecule expression from those that are characteristic of epithelial cells (integrin αvβ3, αωβ5, and E-cadherin) to those of endothelial cells (integrin αvβ3, αωβ5, Platelet Endothelial Cell Adhesion Molecule and Vascular Endothelial-Cadherin), a process referred to as “pseudovasculogenesis” (13,14). In preeclampsia, there is shallow placental cytotrophoblast invasion of uterine spiral arteries, leading to reduced placental perfusion and consequently placental insufficiency. Both in vitro and in vivo studies show that trophoblasts obtained from patients with preeclampsia fail to undergo these alterations of adhesion molecules and pseudovasculogenesis (15,16). The molecular pathways that regulate pseudovasculogenesis may involve a vast array of transcription factors, growth factors, and cytokines (17). Considerable attention has recently been focused on angiogenesis-related gene products such as vascular endothelial growth factor (VEGF), angiotensin/tie, and ephrin family proteins and their role in regulating pseudovasculogenesis and invasiveness. Invasive trophoblasts express VEGF, placental growth factor (PIGF), and VEGF-C and their receptors. Furthermore, blocking their signaling pathways decreases the expression of marker of pseudovasculogenesis in vitro (18).

Generalized endothelial dysfunction may be responsible for all of the clinical aspects of the maternal syndrome in preeclampsia. The identification of circulating factors that mediate endothelial dysfunction has been the source of great research interest for decades. Several groups have reported alterations in cytokines/growth factors/chemicals such as TNF-α, IL-6, IL-1α, IL-1β, Fas ligand, oxidized lipid products, neurokinin-B, and asymmetric dimethyl arginine (ADMA) that are released by the placenta and/or other maternal sources in preeclampsia (18–22). Recently, a lower-than-normal l-arginine concentration that is caused by arginase II overexpression and that may redirect placental endothelial nitric oxide (NO) synthesis toward peroxynitrite was described in preeclampsia (23).
Newer Aspects on Relaxin in Pregnancy and Preeclampsia

Relaxin is produced by the corpus luteum of the ovary and circulates in the luteal phase of the menstrual cycle and rises early in gestation. Human chorionic gonadotrophin produced by the placenta is a major stimulus for relaxin secretion during pregnancy in women. Chronic administration of relaxin to conscious female rats increases GFR and effective renal plasma flow, thereby mimicking the changes in the renal circulation during pregnancy (24,25). This renal vasodilatory effect does not require the presence of the ovaries (25) and is also observed during relaxin administration to male rats (26). Relaxin administration to nonpregnant female rats also diminishes the renal vasoconstrictor response to angiotensin II, similar to the dampening influence of rat gestation (25,27,28). Moreover, reduced myogenic reactivity of small renal arteries is observed after relaxin administration, analogous to vessels isolated from midterm pregnant rats (29,30). Finally, relaxin-neutralizing antibodies or removal of circulating relaxin by ovariectomy completely abolishes the renal vasodilation, hyperfiltration, and reduced myogenic reactivity of small renal arteries, as well as the osmoregulatory changes in midterm pregnant rats (31).

Jeyabalan et al. (32) (Figure 1) recently proposed that relaxin upregulates vascular gelatinase activity during pregnancy, thereby contributing to renal vasodilation, hyperfiltration, and reduced myogenic reactivity of small renal arteries through activation of the endothelial endothelin B (ET_B) receptor–NO pathway. The notion that metalloproteinase 2 (MMP-2) plays a pivotal role in the relaxin-gelatinase pathway is based on the confluence of several observations. First, relaxin, the endothelial ET_B receptor, and NO have an essential role in pregnancy-mediated renal vasodilation. Second, relaxin serves to upregulate MMP expression at least in fibroblasts (33). Third, vascular MMP, such as MMP-2, are able to process big ET at the gly-leu bond to ET_1-32 with subsequent activation of endothelin receptors (34).

By inhibiting gelatinase activity in chronically instrumented rats in vivo and in isolated small renal arteries in vitro, Jeyabalan et al. (32) demonstrated an essential role for vascular gelatinase in the relaxin-mediated, renal circulatory changes of pregnancy. In contrast, the traditional endothelium-converting enzyme pathway that processes big ET to ET_1-21 is not involved on the basis of the lack of any hemodynamic effect of the traditional endothelium-converting enzyme inhibitor phosphoramidon. In small renal arteries harvested from relaxin-treated nonpregnant or midterm pregnant rats, vascular MMP-2 activity is upregulated by ~50%. Thus, vascular gelatinase activity not only is part of the endothelial ET_B-NO vasodilatory pathway but also is a major locus of regulation by relaxin. Finally, relaxin was administered to ET_B receptor–deficient rats. Although the small renal arteries harvested from these rats showed upregulation of vascular MMP-2 activity, they failed to elicit the typical reduction in myogenic reactivity. This observation when taken in the context of the other results (supra vide) indicates that vascular gelatinase is in series with and upstream of the endothelial ET_B-NO signaling pathway in the renal vasodilatory response to pregnancy mediated by relaxin (32).

Circulating levels of immunoreactive H2 relaxin have been reported to be similar in women with preeclampsia and normal pregnancy at comparable gestational ages (35). However, whether circulating relaxin bioactivity may be deficient during the disease is uncertain. The LGR7 and LGR8 relaxin receptors were only recently identified (36). Little is known about their expression on blood vessels. By analogy to other receptor systems, fewer receptors, increased expression of inactive receptors, or soluble receptors could undermine relaxin signaling to the vasculature, thereby compromising renal vasodilation in preeclampsia. Because increased vascular gelatinase activity by relaxin is a proximal step in the vasodilatory pathway of pregnancy, inappropriate MMP-2 activity may contribute to reduced renal function in preeclampsia. Excessive expression of ET or of the ET_A or ET_B receptor on the vascular smooth muscle of renal arterioles may overwhelm the vasodilatory pathway initiated by relaxin. Mutations or polymorphisms of the ET_B receptor or of endothelial NO synthase that reduce activity may predispose a woman to preeclampsia by impairing trophoblast invasion on the one hand (37,38) and by compromising maternal endothelial behavior on the other. Much earlier work indirectly alluded to such possibilities (11).

Circulating Proangiogenic Factors and Their Inhibitors

An increased placental expression and secretion of soluble fms–like tyrosine kinase 1 (sFlt1), a naturally occurring circulating VEGF antagonist in patients with preeclampsia, has recently been demonstrated (39–41). Importantly, when administered exogenously to rats, sFlt1 alone has been shown to

Figure 1. Proposed scheme of cellular mechanisms underlying pregnancy and relaxin-induced renal vasodilation and hyperfiltration and reduced myogenic reactivity of small renal arteries. Big ET is released from Weibel-Palade bodies in response to pulsatile pressure in vivo or increases of intraluminal pressure in vitro. MMP-2, eNOS, and possibly the ET_B receptor are localized in caveolae of endothelial cells.
be sufficient to induce preeclampsia-like phenotype (39). Finally, data using VEGF conditional knockout mice gave us definitive genetic evidence that interference with VEGF signaling in the kidneys leads to clinical preeclampsia, namely proteinuria and glomerular endotheliosis (42).

VEGF is an endothelial-specific mitogen that plays a key role in promoting angiogenesis. VEGF’s activities are mediated primarily by its interaction with two high-affinity receptor tyrosine kinases—kinase-insert domain region (KDR) and Flt1—that are selectively expressed on vascular endothelial cell surface (43). Alternative splicing of Flt1 results in the production of an endogenously secreted protein referred to as sFlt1, which lacks the cytoplasmic and transmembrane domain but retains the ligand-binding domain (44,45) (Figure 2). Thus, sFlt1 can antagonize circulating VEGF by binding to it and preventing VEGF’s interaction with its endogenous receptors. sFlt1 also binds and antagonizes PI GF, another member of the VEGF family that is made in the placenta predominantly.

In vitro studies confirm that excess placental sFlt1 production induces antiangiogenic state in the serum of preeclamptic women that can be rescued by exogenous VEGF and PI GF (39). Excess sFlt1 alone, when administered to pregnant rats, induces albuminuria, hypertension, and renal pathologic changes of glomerular endotheliosis by antagonizing circulating VEGF and PI GF and inducing endothelial dysfunction. In addition, circulating levels of free VEGF and free PI GF were decreased in conjunction with elevated sFlt1 in the bloodstream at the time of disease presentation (39,40). More recently, when free PI GF and free VEGF were measured throughout pregnancy, the growth factors were decreased in preeclamptic women well before the onset of clinical disease (46–48).

Recently, Levine et al. (49) presented exciting evidence suggesting that sFlt1 is a—or perhaps the—circulating factor responsible for preeclampsia. They performed a nested, case-control study within the Calcium for Preeclampsia Prevention Trial that involved healthy nulliparous women. Each woman with preeclampsia was matched to one normotensive control subject. A total of 120 pairs of women were randomly chosen. Serum concentrations of angiogenic factors (total sFlt1, free PI GF, and free VEGF) were measured throughout pregnancy. The sFlt1 level increased beginning ~5 wk before the onset of preeclampsia. At the onset of clinical disease, the mean serum level in the women with preeclampsia was 4382 pg/ml, compared with 1643 pg/ml in control subjects with fetuses of similar gestational age. The PI GF levels were significantly lower in the women who later had preeclampsia than in the control subjects beginning at 13 to 16 wk of gestation (mean, 90 versus 142 pg/ml), with the greatest difference occurring during the weeks before the onset of preeclampsia, coincident with the increase in the sFlt1 level. Alterations in the levels of sFlt1 and free PI GF were greater in women with an earlier onset of preeclampsia and in women in whom preeclampsia was associated with a small-for-gestational-age infant. The authors concluded that increased levels of sFlt1 and reduced levels of PI GF predict the subsequent development of preeclampsia.

These data support the hypothesis that elevated sFlt1 may induce the maternal syndrome and that the elevated sFlt1 is not a consequence of the maternal syndrome. VEGF is known to stimulate angiogenesis, as well as promote vasodilation by stimulating NO and prostacyclin formation, signaling molecules that are decreased in preeclampsia (50). Furthermore, a significant percentage of cancer patients who receive VEGF signaling antagonists develop hypertension and proteinuria (51,52). Even the loss of a single VEGF allele from the glomerulus in genetically modified mice resulted in glomerular endotheliosis and proteinuria (42). It is interesting that dramatic endothelial defects were observed even though the circulating levels of VEGF were unaffected, which emphasizes that tight local regulation of VEGF signaling seems to be critical for endothelial function. These observations suggest that excess sFlt1 may play a causal role in the pathogenesis of the maternal syndrome in preeclampsia by neutralizing VEGF and PI GF (Figure 3). However, there are limitations and several unanswered questions to the sFlt1 story. The precise mechanisms of excess sFlt1 production by the placenta are not known and, importantly, the role of sFlt1 in normal placental development and in placental pseudovascularogenesis is not clear. No coagulation or liver function abnormalities or brain abnormalities ( eclampsia) were reported in sFlt1-treated animals. The mechanisms of proteinuria during VEGF-deficient states are still unclear. Additional synergistic factors that are elaborated by the placenta may yet be identified to play a role in the pathogenesis of the generalized endothelial dysfunction noted in preeclampsia.

**Endothelium-Derived Relaxing Factors and Their Inhibitors**

Endothelial factors such as prostaglandins (PG) and NO have been proposed as mediators of gestational renal vasodilation and hyperfiltration. However, studies in gravid humans and animal models have not shown a compelling role for vasodilatory PG in the pregnancy-induced increases in GFR and effective renal plasma flow (27,53–55) or in the decrease of total peripheral vascular resistance (56,57). The first indi-
cation of a potential role for NO came from the evaluation of guanosine 3',5'-cyclic monophosphate (cGMP), an important second messenger of NO. Plasma concentration, urinary excretion, and metabolic production of cGMP are increased in rat pregnancy and pseudopregnancy (58,59), as well as in human gestation (60–62). Urinary excretion of nitrate and nitrite, stable metabolites of NO, are increased in pregnant and pseudopregnant rats that consume a diet low in nitrite and nitrate corresponding to the rise in cGMP excretion (59). Furthermore, NO metabolites are increased in the plasma of pregnant rats, and NO-hemoglobin in red blood cells is detected in pregnant but not in nonpregnant rats (61). These data suggest that endogenous NO production is increased in gravid rats, although the tissue source(s) of the increased NO metabolites and cGMP remain unknown (63,64).

Specific defects in resistance artery endothelium from women with preeclampsia have been shown (65). Supportive evidence for NO deficiency in preeclampsia has recently been obtained from the reduced uterine perfusion pressure rat model (66). The investigators found that supplementation with L-arginine decreased BP by 19 mmHg in pregnant rats with reduced uterine perfusion pressure (untreated versus treated) as compared with 12 mmHg in pregnant (untreated versus treated) rats. Thus, these results suggest that L-arginine supplementation may be beneficial in attenuating the hypertension in preeclampsia.

An attractive preeclampsia factor that could directly interfere with NO and induce endothelial dysfunction in pregnant women is ADMA. Savvidou et al. (22) tested the hypothesis that ADMA, an endogenous endothelial NO synthase inhibitor, contributes to the development of preeclampsia. A role for NO—or its absence—has been established in earlier studies of preeclampsia. The authors measured forearm ischemia-reperfusion as a marker of endothelial function. They also monitored uterine blood flow by means of Doppler techniques. They searched for the occurrence of intrauterine growth retardation, and they obviously measured ADMA and its symmetrical analog. The authors found that women with evidence for impaired placental perfusion had >30% prevalence of children with intrauterine growth retardation and >20% prevalence of preeclampsia. Women with preeclampsia clearly had significantly lower flow-mediated vasodilation than women with normal uterine perfusion. In women with preeclampsia, there was a remarkably tight correlation between ADMA levels and flow-mediated vasodilation. Taken together, the authors found that endothelial dysfunction develops before preeclampsia, women with higher uterine flow resistances are at risk for intrauterine growth retardation and preeclampsia, and ADMA may be a potentially contributing factor to endothelial dysfunction in these women.

Circulating Autoantibodies

Haller et al. (65,66) observed that a circulating factor in preeclamptic women caused endothelial cells to express surface adhesion molecules and made endothelial cell layers more permeable. The latter process involved the activation of protein kinase C. Wallukat et al. (67) subsequently identified circulating autoantibodies that are capable of activating the angiotensin II (Ang II) AT1 receptor. The autoantibodies (AT-1AA) arise approximately at the time that symptoms develop, namely after the 20th week of gestation, and subside within 6 wk after delivery. AT-1AA were purified and identified as belonging to a fraction of IgG antibodies. Wallukat et al. (67) showed that AT1-AA bound to a certain seven amino acid sequence on the second extracellular loop of AT1-AA. They documented specificity by means of Western blotting and co-localization studies. Indeed, AT-1AA functioned in a Western blot at least as well as currently available commercial antibodies to the AT-1 receptor. Dechend et al. (68,69) confirmed these findings by relying on co-immunoprecipitation studies. They could not find evidence for calcium signaling or smooth muscle cell contraction resulting from the autoantibodies. However, they were able to show that the autoantibodies initiated a signaling cascade that culminated in transcription factor activation (NF-κB and activator protein-1) and subsequently tissue factor expression.

Indepedently, Zia et al. (70) studied 38 pregnant patients, 20 of whom had severe preeclampsia and 18 of whom were normotensive. IgG was purified from these individuals, and the presence of AT1-AA was determined. Immortalized human trophoblasts were chosen to study plasminogen activator inhibitor 1 production and secretion after treatment with IgG from normotensive and preeclamptic women. Their findings suggest that maternal autoantibody with the ability to activate AT1 receptors may account for two features of preeclampsia: increased plasminogen activator inhibitor 1 production and shallow trophoblast invasion.

Recently, Dechend et al. (69) showed that AT-1AA can cause human trophoblasts or vascular smooth muscle cells to
produce reactive oxygen species (ROS) by activating the NADPH oxidase. Vascular smooth muscle cells from p47 phox gene-disrupted −/− and control +/+ mice were subjected to AT1-AA. By means of DCF fluorescence, both Ang II and AT1-AA produced a strong response. The ROS production was greatly ameliorated by the antioxidant tiron or by using vascular smooth muscle cells lacking p47 phox. In these cells, the NADPH oxidase was not operative. In other studies, the investigators used human trophoblasts and electromobility shift assays with supershifts and showed that the NF-κB units p50 and p65 were activated by either Ang II or AT-AA. Human trophoblasts, as shown by the anticytokeratin 7 marker, also exhibited p22 phox.

AT1-AA have also been found in patients with malignant hypertension (71) and in patients with humoral renal transplant rejection (unpublished observations). Thus, they are not a specific preeclampsia phenomenon. The existence of these antibodies is exciting. However, the field is hampered because detection still relies on a bioassay. Attempts to establish an ELISA have not yet been successful. Thus, confirmatory studies in large populations of women with preeclampsia, as with sFlt1, have not yet been conducted.

It is interesting that AT1 receptor heterodimers are increased in preeclampsia. An AT1 receptor-bradykinin-2 receptor heterodimer with increased Ang II signaling has been described (72). A promising transgenic rat model that relies on human renin and angiotensinogen transgenes has been developed (73). This model features the development of AT1-AA (unpublished observations). AT1-AA could be an epiphenomenon. Nevertheless, there are precedents for the notion that activating antibodies can cause disease. The argument that activating antibodies to the β1 adrenergic receptor can cause cardiomyopathy is compelling (74) (Figure 4).

**Conclusions**

Exciting news has emanated from the preeclampsia research arena recently on all fronts. Compelling basic research is in progress. Exciting clinical trials have been conducted, and more are in progress. Is it possible that these divergent mechanisms interact in some way? Ang II plays some role in angiogenesis. Angiotensin-converting enzyme inhibitors and AT1 receptor blockers seem to be antiangiogenic and decrease microvessel formation (75). VEGF-mediated angiogenesis can be decreased with AT1 receptor blockers (76). ADMA generation can be diminished by angiotensin-converting enzyme inhibitors and by AT1 receptor blockers in patients who receive these drugs (77). How Ang II might stimulate ADMA formation is not clear. ADMA is synthesized from methylated arginine residues in proteins by protein arginine methyltransferases. The compound is metabolized to citrulline by the actions of dimethylarginine dimethylaminohydrolases I and II. The enzymes may be impaired in preeclampsia. Could Ang II act to influence the expression of TGF-β3? This mediator has been implicated in regulating trophoblast outgrowth, invasion, or both and must be downregulated at a crucial point (78).

Could Ang II possibly play a role in upregulating sFlt1? The signal presumably has to do with hypoxia and therefore may involve the hypoxia-inducible factor (HIF) (79). It is interesting that Ang II may increase HIF-1α induction. According to Page et al. (80), Ang II relies on ongoing translation to maintain elevated HIF-1α protein levels. Ang II increases HIF-1α translation by an ROS-dependent activation of the phosphatidylinositol 3-kinase pathway, which acts on the 5′-untranslated region of HIF-1α mRNA. Their results suggest that the nonhypoxic induction of the HIF-1α transcription factor via vasoactive hormones such as Ang II might be important to vascular...
biology. Finally, evidence has recently been presented that HIF transcription factors are overexpressed in preeclamptic placentas (81). In vitro DNA binding activity for HIF-1α was demonstrated in these studies. Flt1 and tyrosine hydroxylase, both of which are equipped with hypoxia response elements, were expressed to a greater degree in preeclamptic compared with normal placentas. Mechanisms that contribute to alternative Flt1 splicing would be elucidative.

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