Endocan, a putative endothelial cell marker, is elevated in preeclampsia, decreased in acute pyelonephritis, and unchanged in other obstetrical syndromes

Henry Adekola¹,², Roberto Romero¹,³,⁴, Piya Chaemsaiithong¹,², Steven J. Korzeniewski¹,²,⁴, Zhong Dong¹,², Lami Yeo¹,², Sonia S. Hassan¹,², and Tinnakorn Chaiworapongsas¹,²

¹Perinatology Research Branch, Program for Perinatal Research and Obstetrics, Division of Intramural Research, Eunice Kennedy Shriver National Institute of Child Health and Human Development, NIH, Bethesda, MD (Detroit, MI), USA, ²Department of Obstetrics and Gynecology, Wayne State University School of Medicine, Detroit, MI, USA, ³Department of Obstetrics and Gynecology, University of Michigan, Ann Arbor, MI, USA, and ⁴Department of Epidemiology and Biostatistics, Michigan State University, East Lansing, MI, USA

Abstract

Objective: Endocan, a dermatan sulphate proteoglycan produced by endothelial cells, is considered a biomarker for endothelial cell activation/dysfunction. Preeclampsia is characterized by systemic vascular inflammation, and endothelial cell activation/dysfunction. Therefore, the objectives of this study were to determine whether: (1) plasma endocan concentrations in preeclampsia differ from those in uncomplicated pregnancies; (2) changes in plasma endocan concentration relate to the severity of preeclampsia, and whether these changes are specific or observed in other obstetrical syndromes such as small-for-gestational age (SGA), fetal death (FD), preterm labor (PTL) or preterm prelabor rupture of membranes (PROM); (3) a correlation exists between plasma concentration of endocan and angiogenic (placental growth factor or PI GF)/anti-angiogenic factors (soluble vascular endothelial growth factor receptor or sVEGFR-1, and soluble endoglin or sEng) among pregnancies complicated by preeclampsia; and (4) plasma endocan concentrations in patients with preeclampsia and acute pyelonephritis (both conditions in which there is endothelial cell activation) differ.

Method: This cross-sectional study included the following groups: (1) uncomplicated pregnancy (n = 130); (2) preeclampsia (n = 102); (3) pregnant women without preeclampsia who delivered an SGA neonate (n = 51); (4) FD (n = 49); (5) acute pyelonephritis (AP; n = 35); (6) spontaneous PTL (n = 75); and (7) preterm PROM (n = 64). Plasma endocan concentrations were determined in all groups, and PI GF, sEng and VEGFR-1 plasma concentrations were measured by ELISA in the preeclampsia group.

Results: (1) Women with preeclampsia had a significantly higher median plasma endocan concentration than those with uncomplicated pregnancies (p = 0.004); (2) among women with preeclampsia, the median plasma endocan concentration did not differ significantly according to disease severity (p = 0.1), abnormal uterine artery Doppler velocimetry (p = 0.7) or whether diagnosis was made before or after 34 weeks gestational age (p = 0.3); (3) plasma endocan concentration in women with preeclampsia correlated positively with plasma anti-angiogenic factor concentrations [sVEGFR-1: Spearman rho 0.34, p = 0.001 and sEng: Spearman rho 0.30, p = 0.003]; (4) pregnancies complicated by acute pyelonephritis with bacteremia had a lower median plasma endocan concentration than pregnancies complicated by acute pyelonephritis without bacteremia (p = 0.004), as well as uncomplicated pregnancies (p = 0.001); and (5) there was no significant difference in the median plasma endocan concentration between uncomplicated pregnancies and those complicated by FD, delivery of an SGA neonate, PTL or preterm PROM (other members of the “great obstetrical syndromes”; each p > 0.05).

Conclusion: Median maternal plasma endocan concentrations were higher in preeclampsia and lower in acute pyelonephritis with bacteremia than in uncomplicated pregnancy. No significant difference was observed in the median plasma endocan concentration between other great obstetrical syndromes. Median maternal plasma endocan concentrations were higher in preeclampsia and lower in acute pyelonephritis with bacteremia than in uncomplicated pregnancy. No significant difference was observed in the median plasma endocan concentration between other great obstetrical syndromes.

Keywords

Endothelial cell activation, endothelial dysfunction, fetal death, preterm labor, small-for-gestational age, soluble endoglin, soluble vascular endothelial growth factor receptor-1

History

Received 30 July 2014
Revised 1 September 2014
Accepted 9 September 2014
Published online 28 October 2014

This work was authored as part of the Contributor’s official duties as an Employee of the United States Government and is therefore a work of the United States Government. In accordance with 17 U.S.C. 105, no copyright protection is available for such works under U.S. Law.

This is an Open Access article distributed under the terms of the Creative Commons Attribution-NonCommercial-NoDerivs License (http://creativecommons.org/licenses/by-nc-nd/4.0/), which permits non-commercial re-use, distribution, and reproduction in any medium, provided the original work is properly cited, and is not altered, transformed, or built upon in any way.

Address for correspondence: Roberto Romero, Perinatology Research Branch, NICHD, NIH, DHHS, Wayne State University/Hutzel Women’s Hospital, 3990 John R, Box 4, Detroit, MI 48201, USA. Tel: +1 313 993 2700. Fax: +1 313 993 2694. E-mail: romeror@mail.nih.gov
obstetrical syndromes and uncomplicated pregnancies. The difference in the direction of change of endocan in preeclampsia and acute pyelonephritis with bacteremia may be consistent with the view that both disease entities differ in pathogenic mechanisms, despite their associations with systemic vascular inflammation and endothelial cell activation/dysfunction.

Introduction

The traditional view of the pathogenesis of preeclampsia is that uteroplacental ischemia induces the production of soluble factors (or toxins) that, when released into the maternal circulation, are responsible for the clinical manifestations of the disease [1–9]. These factors are thought to cause intravascular inflammation [10–16], endothelial cell dysfunction [17–25], increased thrombin generation [26–35] and platelet aggregation [3,28,35–39].

Generalized endothelial cell activation/dysfunction is considered to be central to the pathophysiology of preeclampsia [9,17]. Considerable effort has been made to identify circulating markers of endothelial cell activation/dysfunction in the circulation of normal pregnant women and those with preeclampsia – this has included coagulation factors produced by endothelial cells, such as Von Willebrand factor [40–44], cellular ‘‘cements’’ (e.g. cellular fibronectin, which is also almost exclusively located in the endothelium) [45–56], endothelial cell adhesion molecules (e.g. E-selectin and vascular cell adhesion molecule-1) [57–67] and anti-endothelial cell antibodies [68–70]. However, none of these makers have been proven to be specific to preeclampsia, and can be elevated in other conditions [71–80]. Thus, a major challenge has been the to identify a biomarker specific to endothelial cell activation/dysfunction and preeclampsia.

Endocan, also known as endothelial specific molecule-1 (ESM-1), is a proteoglycan detectable in the circulation that has been proposed to be a new endothelial cell marker. This protein is elevated in serum of patients with sepsis who have endothelial cell activation/dysfunction [81,82]. Similarly, the serum concentrations of endocan are elevated in lung, breast, hepatocellular [83] and renal cancers [84–86] as well as acute myeloid leukemia [87], conditions associated with endothelial activation/dysfunction [72,74,76,88–94].

The objectives of this study were to determine: (1) whether plasma endocan concentrations in PE differ from those of uncomplicated pregnancy; (2) whether changes the plasma endocan concentration relate to the severity of preeclampsia, and whether these changes are specific or observed in other obstetrical syndromes such as small-for-gestational age (SGA), fetal death (FD), preterm labor (PTL) or preterm prelabor rupture of membranes (PROM); (3) if a correlation exists between the plasma concentration of endocan and angiogenic/anti-angiogenic factors among pregnancies complicated by PE; and (4) whether plasma endocan concentrations in patients with preeclampsia and acute pyelonephritis (both conditions in which there is endothelial cell activation) differ.

Materials and methods

Study design

A cross-sectional study was designed to include patients in the following groups: (1) uncomplicated pregnancy ($n = 130$); (2) preeclampsia ($n = 102$); (3) pregnant women without pre-eclampsia or hypertension who delivered a small-for-gestational age neonate ($n = 51$); (4) fetal death ($n = 49$); (5) acute pyelonephritis ($n = 35$); (6) preterm labor with intact membranes ($n = 75$); and (7) preterm PROM ($n = 64$). All participants provided written informed consent for the collection and use of samples for research purposes under the protocols approved by the Institutional Review Boards of Wayne State University and the Eunice Kennedy Shriver National Institute of Child Health and Human Development, National Institutes of Health, Department of Health and Human Services (NICHD/NIH/DHHS).

Clinical definitions

Preeclampsia was defined as new onset hypertension (systolic and/or diastolic blood pressure of $\geq 140$ and/or $\geq 90$ mm Hg) that developed after 20 weeks of gestation, measured on at least two occasions, 4 h to 1 week apart and proteinuria ($\geq 300$ mg in a 24-h urine collection, or two random urine specimens obtained 4 h to 1 week apart containing $\geq 1$ + protein by dipstick) [95]. Severe PE was diagnosed according to criteria proposed by the American Congress of Obstetricians and Gynecologists (ACOG) [95,96]. Early and late-onset preeclampsia was defined as cases diagnosed before and after 34 weeks of gestation, respectively [97].

The uncomplicated pregnancy group comprised of women with: (1) no medical, obstetrical or surgical complications; (2) a singleton gestation; (3) no labor; and (4) a normal term ($\geq 37$ weeks) infant delivered at term whose birth weight was between the 10th and 90th percentile for gestational age [98]. Acute pyelonephritis was diagnosed in the presence of fever (temperature $\geq 38^\circ$C), clinical signs or symptoms of an upper urinary tract infection (e.g. flank pain, costovertebral angle tenderness), pyuria and a positive urine culture [99,100]. An SGA neonate was defined as birth weight $<10$th percentile for gestational age [98]. Fetal death was defined as death of the fetus after 20 weeks of gestation, confirmed by ultrasound. All fetal deaths were unexplained. Spontaneous PTL was defined by the presence of preterm labor leading to preterm delivery. Pre-term PROM was diagnosed as amniorrhaxis in preterm gestations that were followed by preterm delivery.

Maternal plasma concentrations of endocan, placental growth factor, sVEGFR-1 and endoglin

Maternal blood was collected into tubes containing ethylene-diaminetetraacetic acid (EDTA), centrifuged and stored at $-70^\circ$C until assayed. Maternal plasma concentrations of intact endocan were measured with an immunoassay following the manufacturers’ instructions (USCN Life Science Inc., Wuhan, Hubei, PRC or Cloud-Clone Corp., Houston, TX). Maternal plasma concentrations of placenta growth factor (PIGF), soluble endoglin (sEng) and soluble vascular endothelial growth factor receptor (sVEGFR)-1 were determined by sensitive and specific immunoassays obtained from R&D.
Doppler velocimetry

Color Doppler was used to identify blood vessels, and spectral Doppler to calculate Doppler indices in the uterine arteries. The examinations were performed at the time of diagnosis according to methods previously described [15,102,103]. The uterine artery resistance index (RI) was used as a measure of vascular impedance in the uterine circulation. A mean RI (average of left and right) of < or ≥95th percentile for gestational age was used to determine normal and abnormal uterine artery Doppler velocimetry, respectively [104].

Statistical analysis

Normality of data was assessed using the Kolmogorov–Smirnov test and visual plot inspection. The Kruskal–Wallis test with post-hoc analysis by Mann–Whitney U-tests was used to compare continuous variables. Comparison of proportions was performed using χ² or Fisher’s exact tests. Spearman’s rank correlation coefficient was used to assess the relationship between plasma endocan, angiogenic (PIGF) and anti-angiogenic factor [(sEng and sVEGFR)-1] concentrations as well as maternal age, gestational age at blood draw, gestational age at delivery and newborn birth weight. General linear models were constructed to examine the relevance of potential confounders including gestational age at venipuncture, maternal age, African American race and history of smoking status. Endocan concentrations were log-transformed. Multivariable analysis also controlled for the false discovery rate (FDR) in light of the performance of multiple tests. A probability value of <0.05 (2-tailed) was considered significant. Statistical tests were performed with Statistical Package for the Social Sciences version 19 (SPSS Inc., Chicago, IL).

Results

The demographic, clinical and obstetric characteristics of the study population are displayed in Table 2. Endocan was detected in the maternal plasma of all patients. Among women with uncomplicated pregnancies, maternal plasma endocan concentrations did not correlate with maternal age (p = 0.5), gestational age at venipuncture (p = 0.2), gestational age at delivery (p = 0.9) or birth weight of the neonate (p = 0.8).

Maternal plasma endocan concentrations in pre-eclampsia

The median plasma endocan concentration (ng/ml) in patients with preeclampsia was significantly higher than that of women with uncomplicated pregnancies (22.5, IQR 13.8–44.4 versus 18.2, IQR 10.6–28.0; p = 0.004; Figure 1). Subgroup analysis performed among women with preeclampsia revealed that the median plasma endocan concentration (ng/ml) did not significantly differ according to: disease severity (mild preeclampsia 17.5, IQR 10.5–34.1 versus severe preeclampsia 22.6, IQR 15.3–45.6; p = 0.1); the presence of abnormal uterine artery Doppler velocimetry (normal: 21.4, IQR 14.2–51.0 versus abnormal: 22.3, IQR 13.3–41.0; p = 0.7); or if diagnosis was made before or after 34 weeks gestational age (early onset: 24.0, IQR 17.1–45.0 versus late-onset: 22.0, IQR 12.0–43.0; p = 0.3). Maternal plasma endocan concentration correlated positively with plasma sVEGFR-1 (Spearman’s rho 0.34; p = 0.001) and sEng (Spearman’s rho 0.30; p = 0.003), but not with the concentration of PIGF (p = 0.3).

Table 1. Sensitivities and coefficients of variation of the assays used in this study.

| Analytes                        | Sensitivity | Inter-assay coefficient of variation (%) | Intra-assay coefficient of variation (%) |
|---------------------------------|-------------|-----------------------------------------|------------------------------------------|
| Endocan (ng/ml)                 | 89.5        | 6.2                                     | 10.2                                     |
| Soluble endoglin (ng/ml)        | 0.08        | 2.0                                     | 4.0                                      |
| Soluble vascular endothelial growth factor receptor (pg/ml) | 16.97      | 1.4                                     | 3.9                                      |
| Placenta growth factor (pg/ml)  | 9.52        | 6.02                                    | 4.8                                      |

Table 2. Clinical and obstetric characteristics of normal and complicated pregnancies.

|                         | Uncomplicated pregnancy | Pre-eclampsia | SGA | Fetal death | Acute pyelonephritis | PTL | PPROM | p value |
|-------------------------|-------------------------|---------------|-----|-------------|----------------------|-----|-------|---------|
| Age (years)             | 25 (21–29)              | 23.5 (19.8–30)| 24 (20–29)| 26 (20–30) | 22 (19–25)          | 22.5 (19–26) | 26 (21–32) | <0.001 |
| Nulliparity (%)         | 35 (26.9%)              | 63 (61.8%)    | 26 (50.9%)| 19 (38.8%)  | 12 (34.3%)           | 39 (40.6%) | 17 (26.6%) | <0.001 |
| Race                    |                         |               |     |             |                      |     |       |         |
| African American        | 102 (78.5%)             | 83 (81.4%)    | 44 (86.3%)| 42 (85.7%)  | 27 (77.1%)           | 60 (80%) | 57 (89.1%) | 0.55    |
| Caucasian               | 15 (11.5%)              | 11 (10.8%)    | 4 (7.8%)  | 3 (6.1%)    | 5 (14.3%)            | 9 (12.3%) | 6 (9.4%)  |         |
| Hispanic                | 7 (5.4%)                | 5 (4.9%)      | 1 (2.0%)  | 3 (6.1%)    | 3 (8.6%)             | 3 (4%)   | 1 (1.6%)  |         |
| Others                  | 6 (4.6%)                | 3 (2.9%)      | 2 (3.9%)  | 1 (2%)      | 0                    | 1 (1.3%) | 0       |         |
| Smoking                 | 22 (16.9%)              | 14 (13.7%)    | 15 (29.4%)| 16 (32.7%)  | 5 (14.3%)            | 27 (28.1%)| 34 (53.1%)| <0.001  |
| GA at venipuncture      | 38                      | 36.1          | 36.9     | 31          | 31.4                 | 29.9    | 30.6    | <0.001  |
| (weeks)                 | (31.4–39.1)             | (31.5–38.6)   | (32.7–38.4)| (24.8–36.6)| (25.3–36.4)          | (25.1–32.3)| (27.6–32.1)|        |
| GA at delivery          | 39.3                    | 36.1          | 37.1     | 31          | 39.4                 | 30      | 31.6    | <0.001  |
| (weeks)                 | (38.4–40.3)             | (32.3–38.6)   | (33.6–38.6)| (25.9–36.7)| (38.4–40.7)          | (25–34) | (29.3–33.1)|        |
| Birth weight (g)        | 3352                    | 2280          | 2050     | 1380        | 3210                 | 1785    | 1580    | <0.001  |
|                         | (3118–3633)             | (1455–2835)   | (1500–2380)| (535–2263) | (2690–3600)          | (865–2623.8)| (1142–2055)|        |

Data presented as median (interquartile range) or number (percentage). GA, gestational age; SGA, small for gestational age; PTL, spontaneous preterm labor with intact membranes; PPROM, preterm prelabor rupture of membranes.
Maternal plasma endocan concentration in pregnancies with acute pyelonephritis

The median plasma endocan concentration (ng/ml) was lower in pregnancies complicated by acute pyelonephritis than in uncomplicated pregnancies, but this was not statistically significant (13.4, IQR 8.3–29.6 versus 18.2, IQR 10.6–28.0; p = 0.2). There was no significant difference observed between the median plasma endocan concentration (ng/ml) in pregnancies complicated by acute pyelonephritis without bacteremia and uncomplicated pregnancies (18.1, IQR 9.5–35.2 versus 18.2, IQR 10.6–28.0; p = 0.7; Figure 2). The median plasma concentration of endocan (ng/ml) in pregnancies complicated by acute pyelonephritis with bacteremia was significantly lower than that of those without bacteremia (8.4, IQR 4.5–13.8 versus 18.1, IQR 9.5–35.2; p = 0.004; Figure 2) and lower than that of women with uncomplicated pregnancies (8.4, IQR 4.5–13.8 versus 18.2, IQR 10.6–28.0; p = 0.001). The prevalence of acute respiratory distress syndrome (ARDS) among pregnancies complicated by acute pyelonephritis in this study was 2.9% (1/35). The plasma endocan concentration of the patient who developed ARDS was 13.4 ng/ml.

The median maternal plasma concentration of endocan (ng/ml) was significantly lower in patients with acute pyelonephritis in patients with preeclampsia (13.4, IQR 8.3–29.6 versus 22.5, IQR 13.8–44.4; p = 0.005).

Maternal plasma endocan concentration in fetal death, SGA, preterm labor and preterm PROM

There were no significant differences in the median plasma endocan concentrations (ng/ml) among women with uncomplicated pregnancies (18.2, IQR 10.6–28.0) and those with FD (19.5, IQR 11.3–39.2; p = 0.3), delivery of an SGA neonate (19.8, IQR 10.8–28.8; p = 0.4); fetal death (19.5, IQR 11.3–39.2; p = 0.3); acute pyelonephritis (13.4, IQR 8.3–29.6; p = 0.2); preterm labor (17.4, IQR 11.8–29.7; p = 0.7); and preterm PROM (15.8, IQR 10.0–26.0; p = 0.5).

Multivariable adjustment with correction for the FDR did not alter the unadjusted determinations of statistical significance. Log endocan concentrations were significantly higher among women with preeclampsia than in those with uncomplicated pregnancies (p < 0.01), adjusting for gestational age at venipuncture, maternal age, nulliparity, race, smoking status and BMI. Similarly adjusting for potential confounders, log endocan concentrations were significantly lower among women with acute pyelonephritis with bacteremia than among those with acute pyelonephritis without bacteremia (p < 0.01) or uncomplicated pregnancies (p < 0.01).

Discussion

Principal findings of the study

(1) The median plasma endocan concentration was higher in preeclampsia than in uncomplicated pregnancies; however,
there was no relationship between the plasma concentration of endocan and the severity of preeclampsia; (2) there was a positive correlation between the plasma concentration of endocan and sVEGFR-1 and sEng plasma concentrations among women with preeclampsia; (3) contrary to what was expected, the median plasma concentration of endocan was lower in patients with acute pyelonephritis than in those with preeclampsia; (4) endocan was not elevated in other obstetrical syndromes, such as fetal death, SGA, preterm labor and preterm PROM, suggesting that an elevation of maternal plasma endocan occurs selectively in patients with preeclampsia.

Endothelial cell activation and dysfunction in health and disease

The endothelium is a monolayer that lines all blood vessels. The functions of the endothelium are to maintain vascular tone, prevent cell adhesion, promote thromboresistance and regulate smooth muscle vessel wall proliferation [105,106]. Many of these properties are mediated by nitric oxide (NO), which is produced by the endothelium [from L-arginine by the action of endothelial NO synthase (also called eNOS)] [107–116]. This gas diffuses to the vascular smooth muscle cells and activates guanylate cyclase, which leads to cGMP-mediated vasodilation [106,107,117]. Physiologic regulators of eNOS expression include shear stress [118–122], but other factors can activate this enzyme, including bradykinin, adenosine, vascular endothelial growth factor and serotonin [112,123–128].

The concept of endothelial cell activation was formulated by investigators examining the behavior of endothelial cells in culture, and was coined to describe the increased adhesive properties of these cells to white blood cells when endothelial cells are exposed to biomechanical stimuli [129–131] or cytokines [132–138]. The molecular basis for the increased adhesiveness was the expression of cell surface adhesion molecules, such as VCAM-1, ICAM-1 and endothelial cell adhesion molecule (also known as E-selectin or ELAM) [139,140]. Nitric oxide generated from endothelium (as a result of the activity of nitric oxide synthase) can reduce endothelial cell activation through inhibition of NFκ-B [139,141–143].

The term ‘endothelial cell dysfunction’ was introduced by physiologists and cardiologists who were originally studying impaired endothelial cell-dependent relaxation, and demonstrated that this feature was present in patients with essential hypertension [144]. Endothelial cell dysfunction has been defined as decreased synthesis, release and/or activity of endothelium-derived nitric oxide induced by hypercholesterolemia [140,145], smoking [140,146–148] or oxidative stress [140,149–152].

Endothelial cell activation may lead to endothelial cell dysfunction [140], and both can induce vasoconstriction, platelet aggregation, leukocyte adhesion, low-density lipoprotein oxidation and matrix metalloproteinase protein activation [140]. Thus, endothelial cell activation/dysfunction can lead to atherosclerosis/vascular disease [153–156]. Pregnancy represents a physiological state in which there appears to be endothelial cell activation as a consequence of physiologic intravascular inflammation [157]. It is unclear if endothelial cell activation and dysfunction have different molecular fingerprints; indeed, they appear to coexist. Preeclampsia is considered to be characterized by endothelial cell activation/dysfunction [17–24], and this condition is associated with a significant increase in the maternal circulating concentrations of sE-selectin [57,58,60,158,159] and sVCAM-1 [57,60,158,160,161], which are markers of endothelial cell activation/dysfunction [136,162]. However, there is no increase in the concentration of sICAM-1 and sPECAM-1 [57,60,163,164], indicating that there are some unique features of endothelial cell activation/dysfunction in preeclampsia. When assessing the profile of adhesion molecules, we have found that patients with acute pyelonephritis have
an increase in sICAM-1, sE-selectin and sVCAM-1 [60], suggesting that there are subtle differences in the adhesion molecule profile in patients with pyelonephritis and pre-eclampsia, both of which are characterized by intravascular inflammation [12,157].

What is endocan?
Endocan is a dermatan sulphate proteoglycan first isolated from the human umbilical vein endothelial cell (HUVEC) cDNA library by Lassalle et al. [165]. This protein is found in endothelial cells and in the epithelium the lung and kidney [166]. Pro-inflammatory cytokines such as tumor necrosis factor (TNF-α) and interleukin (IL)-1β can up-regulate mRNA expression of endocan in endothelial cells [167]. Endocan can inhibit the interaction between intercellular adhesion molecule-1 (ICAM-1) and the integrin (lymphocyte function-associated antigen-1) LFA-1 on leukocytes [168,169], then modulate several leukocyte functions, including adhesion to the endothelium and transmigration [168,170,171]. In addition, endocan can stimulate endothelial cell proliferation and migration induced by epidermal growth factor (EGF), Hepatocyte growth factor/scatter factor (HGF/SF) and vascular endothelial growth factor (VEGF) A and-C [172–174]. This has been attributed to the dermatan sulfate moieties of endocan [172,173,175].

We decided to study the behavior of endocan in normal pregnancy and pregnancy complications due to the claim that this was an endothelial cell marker [81]. Previous reports indicated that serum endocan concentrations were elevated in patients with sepsis and septic shock [81]. Sepsis is considered to represent a state in which there is endothelial cell activation/dysfunction. In these conditions, activated leukocytes roll, adhere and extravasate following interaction between the integrin leukocyte function-associated antigen (LFA-1) and intercellular adhesion molecule (ICAM-1) on surface of these activated leukocytes [176].

Plasma endocan concentration is increased in pre-eclampsia
‘We found that plasma endocan concentrations was increased in patients with preeclampsia, and this increase correlated with the increase in plasma anti-angiogenic factors concentrations, but not with the severity of disease. Preeclampsia is characterized by excessive maternal systemic vascular inflammation, as demonstrated by the phenotypic and metabolic characteristics of neutrophils and monocytes [10,12,177,178], as well as the increased concentration of cytokines [63,65,179–197], chemokines [63,65,198–203], other inflammatory mediators [63,185,189,190,192,204–213], as well as acute phase protein reactants [193,204,214–225] and the decreased concentration of negative acute phase protein reactants [225,226]. The increased cytokine concentration in preeclampsia may be responsible for the elevation in endocan.

The relationship between the increased concentrations of maternal plasma endocan and that of anti-angiogenic factors (sVEGFR-1 and endoglin) in preeclampsia suggests that there may be convergence of the inflammatory process, and the abnormal anti-angiogenic profile observed in the disease [227]. The lack of correlation between the concentrations of PIGF and endocan is unexpected, given that VEGF (another angiogenic factor in the same family as PIGF) can stimulate endocan mRNA expression and release from endothelial cells [228].

Plasma endocan concentration is lower in pregnant women with acute pyelonephritis
Scherpereel et al. [81] demonstrated that the serum endocan concentrations in patients with sepsis and a systemic inflammatory response were significantly higher than that of non-pregnant patients. Our findings herein are different, as we observed that the median plasma endocan concentration was lower in pregnancies complicated by acute pyelonephritis than in non-pregnant patients. This is a puzzling observation, given that our systematic studies of the behavior of cytokines [229], chemokines [230], complement [231,232] in acute pyelonephritis and preeclampsia suggest that both conditions are associated with a pro-inflammatory state. However, studies of the transcriptome of peripheral blood in patients with preeclampsia [233] and pyelonephritis [234] suggest that the molecular details of the inflammatory response differ. Further work is required to understand the similarities and differences in the systemic and local inflammatory response in these two conditions. Interestingly, systemic infection in non-pregnant subjects is associated with an increase in the concentration of sVEGFR-1 in non-pregnant animals [235] and humans [236]. However, in pregnant subjects with acute pyelonephritis, the median plasma concentrations of anti-angiogenic factors (VEGFR-1 and sEng), similar to endocan, are not significantly higher than than that of uncomplicated pregnancy [229]. In addition, endocan behaves in a different direction of change in preeclampsia compared to the changes observed in acute pyelonephritis with bacteremia. This is consistent with the view that there may be a fundamental difference in the nature of the inflammatory response in microbial- and ‘danger signal’-induced inflammation [237].

TNF-α, which stimulates the production of endocan [167], is increased in the peripheral blood of patients with sepsis [238–241]. In addition to demonstrating a high concentration of TNF-α in plasma during maternal sepsis [242], our group also showed a higher concentration of this cytokine in pregnancies complicated by acute pyelonephritis than uncomplicated pregnancies [229]. Therefore, a lower plasma endocan concentration in pregnancies complicated by acute pyelonephritis as a whole, and especially those complicated by bacteremia, was unexpected.

One study reported that, among patients with major trauma, those with lower circulating concentrations of endocan are at increased risk for acute lung injury supporting a protective effect of this protein [243]. The presence of endocan may inhibit leukocyte recruitment, and this protects against lung injury [168]. There was no evidence in our study that patients with pyelonephritis had a higher rate of acute lung injury, although it is well-known that pregnant women with pyelonephritis are at an increased risk for ARDS [244–249].
Strengths and limitations

This is the first study to focus on the changes in plasma concentrations of endocan in preeclampsia, acute pyelonephritis and other “great obstetrical syndromes”. The cross-sectional nature of this study does not enable us to make inferences about temporal changes before diagnosis of the diseases.

Conclusion

Maternal plasma endocan concentrations were higher in pregnancies complicated by preeclampsia and lower in pregnancies complicated by acute pyelonephritis with bacteremia when compared to uncomplicated pregnancies. Patients with SGA, fetal death, preterm labor or preterm PROM did not have demonstrable changes in maternal plasma concentrations of endocan when compared to uncomplicated pregnancies.

Declaration of interest

The authors declare no conflicts of interest. This research was supported, in part, by the Perinatology Research Branch, Division of Intramural Research, Eunice Kennedy Shriver National Institute of Child Health and Human Development, National Institutes of Health, Department of Health and Human Services (NICHD/NIH); and, in part, with Federal funds from NICHD, NIH under Contract No. HHSN275201300006C.

References

1. Young J. The aetiology of eclampsia and albuminuria and their relation to accidental haemorrhage: an anatomical and experimental investigation. Proc R Soc Med 1914;7:307–48.
2. Page EW. On the pathogenesis of pre-eclampsia and eclampsia. J Obstet Gynaecol Br Commomw 1972;79:883–94.
3. Romero R, Lockwood C, Oyarzun E, et al. Toxemia: new concepts in an old disease. Semin Perinatol 1988;12:302–23.
4. Redman CW, Sargent IL. Latest advances in understanding preeclampsia. Science 2005;308:1592–4.
5. Roberts JM, Gammill HS. Preeclampsia: recent insights. Hypertension 2005;46:1243–9.
6. Sibai B, Dekker G, Kupferminc M. Pre-eclampsia. Lancet 2005;365:785–99.
7. Lindheimer MD, Roberts JM, Cunningham GC, et al. Introduction, history, controversies and definitions. In: Lindheimer MD, Roberts JM, Cunningham GC, eds. Chesley’s hypertensive disorders in pregnancy. San Diego: Elsevier;2009:1–24.
8. Steegers EA, von Dadelszen P, Duvekot JJ, et al. Pre-eclampsia. Lancet 2010;376:631–44.
9. Chaiworapongs T, Chaemsaihong P, Yeo L, et al. Pre-eclampsia part 1: current understanding of its pathophysiology. Nat Rev Nephrol 2014;10:466–80.
10. Sacks GP, Studena K, Sargent K, et al. Normal pregnancy and preeclampsia both produce inflammatory changes in peripheral blood leukocytes akin to those of sepsis. Am J Obstet Gynecol 1998;179:80–6.
11. Redman CW, Sacks GP, Sargent IL. Preeclampsia: an excessive maternal inflammatory response to pregnancy. Am J Obstet Gynecol 1999;180:499–506.
12. Gervasi MT, Chaiworapongs T, Pacora P, et al. Phenotypic and metabolic characteristics of monocytes and granulocytes in preeclampsia. Am J Obstet Gynecol 2001;185:792–7.
13. Redman CW, Sargent IL. Pre-eclampsia, the placenta and the maternal systemic inflammatory response – a review. Placenta 2003;24:S21–7.
14. Redman CW, Sargent IL. Preeclampsia and the systemic inflammatory response. Semin Nephrol 2004;24:565–70.
15. Chaiworapongs T, Romero R, Korzeniewski SJ, et al. Maternal plasma concentrations of angiogenic/antiangiogenic factors in the third trimester of pregnancy to identify the patient at risk for stillbirth at or near term and severe late preeclampsia. Am J Obstet Gynecol 2013;208:287.e1–15.
16. Bouwland-Both MI, Steegers EA, Lindemans J, et al. Maternal soluble fms-like tyrosine kinase-1, placent growth factor, plasminogen activator inhibitor-2, and folate concentrations and early fetal size: the Generation R study. Am J Obstet Gynecol 2013;209:121.e1–11.
17. Roberts JM, Taylor RN, Musci TI, et al. Preeclampsia: an endothelial cell disorder. Am J Obstet Gynecol 1989;161:1200–4.
18. Roberts JM, Taylor RN, Goldfien A. Clinical and biochemical evidence of endothelial cell dysfunction in the pregnancy syndrome preeclampsia. Am J Hypertens 1991;4:700–8.
19. Friedman SA, Schiff E, Emeis JJ, et al. Biochemical corroboration of endothelial involvement in severe preeclampsia. Am J Obstet Gynecol 1995;172:202–3.
20. Lyall F, Greer IA. The vascular endothelium in normal pregnancy and pre-eclampsia. Rev Reprod 1996;1:107–16.
21. Roberts JM. Endothelial dysfunction in preeclampsia. Semin Reprod Endocrinol 1998;16:5–15.
22. Taylor RN, de Groot CJ, Cho YK, et al. Circulating factors as markers and mediators of endothelial cell dysfunction in pre-eclampsia. Semin Reprod Endocrinol 1998;16:17–31.
23. Cindrova-Davies T. Gabor than award lecture 2008: pre-eclampsia – from placental oxidative stress to maternal endothelial dysfunction. Placenta 2009;30:555–65.
24. Lamarca B. Endothelial dysfunction. An important mediator in the pathophysiology of hypertension during pre-eclampsia. Minerva Ginecol 2012;64:309–20.
25. Sandvik MK, Leirul G, Nygard O, et al. Preeclampsia in healthy women and endothelial dysfunction 10 years later. Am J Obstet Gynecol 2013;209:569.e1–10.
26. Cunningham FG, Pritchard JA. Hematologic considerations of pregnancy-induced hypertension. Semin Perinatol 1978;2:29–38.
27. Weenink GH, Trefters PE, Vijn P, et al. Antithrombin III levels in preeclampsia correlate with maternal and fetal morbidity. Am J Obstet Gynecol 1984;148:1092–7.
28. Romero R, Mazor M, Lockwood CJ, et al. Clinical significance, prevalence, and natural history of thrombocytopenia in pregnancy-induced hypertension. Am J Perinatol 1989;6:32–8.
29. de Boer K, ten Cate JW, Sturk A, et al. Enhanced thrombin generation in normal and hypertensive pregnancy. Am J Obstet Gynecol 1989;160:95–100.
30. Cadroy Y, Grandjean H, Pichon J, et al. Evaluation of six markers of haemostatic system in normal pregnancy and pregnancy complicated by hypertension or pre-eclampsia. Br J Obstet Gynaecol 1993;100:416–20.
31. Chaiworapongs T, Yoshimatsu J, Espinoza J, et al. Evidence of in vivo generation of thrombin in patients with small-for-gestational-age fetuses and pre-eclampsia. J Matern Fetal Neonatal Med 2002;11:362–7.
32. Dekker G. Prothrombotic mechanisms in preeclampsia. Throm Res 2005;115:17–21.
33. Erez O, Romero R, Kim SS, et al. Over-expression of the thrombin receptor (PAR-1) in the placenta in preeclampsia: a mechanism for the intersection of coagulation and inflammation. J Matern Fetal Neonatal Med 2008;21:345–55.
34. Erez O, Romero R, Hoppensteadt D, et al. Tissue factor and its natural inhibitor in pre-eclampsia and SGA. J Matern Fetal Neonatal Med 2008;21:855–69.
35. Kenny LC, Baker PN, Cunningham FG. Platelets, coagulation, and the liver. In: Lindheimer MD, Roberts JM, Cunningham GC, eds. Chesley’s hypertensive disorders in pregnancy. San Diego: Elsevier;2009:335–51.
36. Socol ML, Weiner CP, Louis G, et al. Platelet activation in preeclampsia. Am J Obstet Gynecol 1985;151:494–7.
37. Cuijpers P, Deutinger J, Tatra G. Platelet specific proteins (beta-thromboglobulin and platelet factor 4) in normal pregnancy and in pregnancy complicated by preeclampsia. Arch Gynecol Obstet 1989;244:91–5.
38. Ahmed Y, van Iddekinge B, Paul C, et al. Retrospective analysis of platelet numbers and volumes in normal pregnancy and in pre-eclampsia. Br J Obstet Gynaecol 1993;100:216–20.
39. Major HD, Campbell RA, Silver RM, et al. Synthesis of sFlt-1 by platelet-monocyte aggregates contributes to the pathogenesis of preeclampsia. Am J Obstet Gynecol 2014;210:547.e1–7.

40. Deng L, Bremke K, Hansson LO, et al. Plasma levels of von Willebrand factor and fibronectin as markers of persisting endothelial damage in preeclampsia. Obstet Gynecol 1994;84:941–5.

41. Molvarec A, Rigo Jr J, Boze T, et al. Increased plasma von Willebrand factor antigen levels but normal von Willebrand factor cleaving protease (ADAMTS13) activity in preeclampsia. Thromb Haemost 2009;101:305–11.

42. Stepanian A, Aybek M, Sanglier T, et al. Von Willebrand factor and ADAMTS13: a candidate couple for preeclampsia pathophysiology. Arterioscler Thromb Vasc Biol 2011;31:1703–9.

43. Xiong Y, Zhou SF, Zhou R, et al. Alternations of maternal and cord plasma hemostasis in preeclampsia before and after delivery. Hypertens Pregnancy 2011;30:347–58.

44. Aref S, Goda H. Increased VWF antigen levels and decreased ADAMTS13 activity in preeclampsia. Hematology 2013;18:237–41.

45. Stubbs TM, Lazarchick J, Horger III EO. Plasma fibronectin levels in preeclampsia: a possible biochemical marker for vascular endothelial damage. Am J Obstet Gynecol 1984;150:885–7.

46. Friedman SA, de Groot CJ, Taylor RN, et al. Plasma cellular fibronectin as a measure of endothelial involvement in preeclampsia and intrauterine growth retardation. Am J Obstet Gynecol 1994;170:838–41.

47. Kupferminc MJ, Peaceman AM, Wigton TR, et al. Fetal fibronectin levels are elevated in maternal plasma and amniotic fluid of patients with severe preeclampsia. Am J Obstet Gynecol 1995;172:649–53.

48. Gredmark T, Bergman B, Hellstrom L. Total fibronectin in maternal plasma as a predictor for preeclampsia. Gynecol Obstet Invest 1999;47:89–94.

49. Chavarria ME, Lara-Gonzalez L, Gonzalez-Gleason A, et al. Maternal plasma cellular fibronectin concentrations in normal and preeclamptic pregnancies: a longitudinal study for early prediction of preeclampsia. Am J Obstet Gynecol 2002;187:595–601.

50. Aydin T, Varol FG, Sayin NC. Third trimester maternal plasma total fibronectin levels in pregnancy-induced hypertension: results of a tertiary center. Clin Appl Thromb Hemost 2006;12:33–9.

51. Leeflang MM, Crossen JS, van der Post JA, et al. Accuracy of fibronectin tests for the prediction of pre-eclampsia: a systematic review. Eur J Obstet Gynecol Reprod Biol 2007;133:12–9.

52. Powers RW, Catov JM, Bodnar LM, et al. Evidence of endothelial dysfunction in preeclampsia and risk of adverse pregnancy outcome. Reprod Sci 2008;15:374–81.

53. Dane C, Buyukasik H, Dane B, et al. Maternal plasma fibronectin and advanced oxidative protein products for the prediction of preeclampsia in high risk pregnancies: a prospective cohort study. Fetal Diagn Ther 2009;26:189–94.

54. Uzun H, Konukoglu D, Albayrak M, et al. Increased maternal plasma fibronectin as a measure of endothelial involvement in preeclampsia. J Obstet Gynecol Reprod Biol 2010;12:573–9.

55. Metz TD, Allshouse AA, Euser AG, et al. Preeclampsia in high risk women is characterized by risk group-specific abnormalities in serum biomarkers. Am J Obstet Gynecol 2014; [Epub ahead of print].

56. Rappaport VI, Hirata G, Yap HK, et al. Anti-vascular endothelial cell antibodies in severe preeclampsia. Am J Obstet Gynecol 1990;162:138–46.

57. Yamamoto T, Takahashi Y, Kuno S, et al. Effects of anti-endothelial cell antibody in pre-eclampsia on endothelin-1 release from cultured endothelial cells. Immunol Cell Biol 1997;75:340–4.

58. Yamamoto T, Geshi Y, Kuno S, et al. Anti-endothelial cell antibody in preeclampsia: clinical findings and serum cytotoxicity to endothelial cell. Nihon Rinsho Meneki Gakkai Kaishi 1998;21:191–7.

59. Rubin DB, Wiener-Kronish JP, Murray JF, et al. Elevated von Willebrand factor antigen is an early plasma predictor of acute lung injury in nonpulmonary sepsis syndrome. J Clin Invest 1990;86:474–80.

60. Sudhoff T, Wehmeier A, Kliche KO, et al. Levels of circulating endothelial adhesion molecules (sE-selectin and sVCAM-1) in adult patients with acute leukemia. Leukemia 1996;10:682–6.

61. Kayal S, Jais JP, Aguiní N, et al. Elevated circulating E-selectin, intercellular adhesion molecule 1, and von Willebrand factor in patients with severe infection. Am J Respir Crit Care Med 1998;157:776–84.

62. McGill SN, Ahmed NA, Christou NV. Increased plasma von Willebrand factor in the systemic inflammatory response syndrome is derived from generalized endothelial cell activation. Crit Care Med 1998;26:296–300.

63. Lyall F, Hayman RG, Ashworth JR, et al. Relationship of cell adhesion molecule expression to endothelium-dependent relaxation in normal pregnancy and pregnancies complicated with preeclampsia or fetal growth restriction. J Soc Gynecol Invest 1999:6:196–201.

64. Bruserud O, Ulvestad E. Expression and release of adhesion molecules by human acute myelogenous leukemia blasts. Leuk Lymphoma 1993:23:149–57.

65. Bretelle F, Sabatier F, Blann A, et al. Maternal endothelial soluble cell adhesion molecules with isolated small for gestational age fetuses: comparison with pre-eclampsia. BJOG 2001;108:1277–82.

66. Parra-Cordero M, Turan OM, Kaur A, et al. Maternal serum soluble adhesion molecule levels at 11 + 0–13 + 6 weeks and subsequent development of preeclampsia. J Matern Fetal Neonatal Med 2007;20:793–6.

67. Costa C, Touscoz GA, Bergallo M, et al. Non-organ-specific and maternal serum soluble adhesion molecules in evaluating endothelial cell activation in preeclampsia. J Matern Fetal Neonatal Med 2015;28(14): 1621–1632.
Scherperel A, Depontieu F, Grigoriu B, et al. Endocan, a new endothelial marker in human sepsis. Crit Care Med 2006;34:532–7.

De Freitas Caires N, Legendre B, Parmentier E, et al. Identification of a 14 kDa endothelial fragment generated by cathepsin G, a novel circulating biomarker in patients with sepsis. J Pharm Biomed Anal 2013;78–79:49–51.

Ozaki K, Toshikuni N, George J, et al. Serum endocan as a novel prognostic biomarker in patients with hepatocellular carcinoma. J Cancer 2014;5:221–30.

van T Veer LJ, Dai H, van de Vijver MJ, et al. Gene expression profiling predicts clinical outcome of breast cancer. Nature 2002;415:530–6.

Lenburg ME, Liou LS, Gerry NP, et al. Previously unidentified changes in renal cell carcinoma gene expression identified by parametric analysis of microarray data. BMC Cancer 2003;3:31.

Borzuk AC, Shah L, Pearson GD, et al. Molecular signatures in biopsy specimens of lung cancer. Am J Respir Crit Care Med 2004;170:167–74.

Hatfield KJ, Lassalle P, Leiva RA, et al. Serum levels of endothelium-derived endocan are increased in patients with severe and mild preeclampsia. J Matern Fetal Neonatal Med 2010;23:820–7.

Favaro D, Santarosa M, Quaia M, et al. Interleukin-6 and soluble intercellular adhesion molecule-1 in renal cancer patients and cultured renal cancer cells. Urol Oncol 1997;3:51–8.

Hoffmann R, Franzke A, Buer J, et al. Prognostic impact of in vivo soluble cell adhesion molecules in metastatic renal cell carcinoma. Br J Cancer 1999;79:1742–5.

Zhang WH, Qiao ZH, Fan XH, et al. The role of intercellular adhesion molecule-1 in binding of acute myeloid leukemic blasts cells to human umbilical vein endothelial cells. Zhonghua Nei Ke Za Zhi 2003;42:413–6.

Basoglu M, Atamanalp SS, Yildirgin MI, et al. Correlation between the serum values of soluble intercellular adhesion molecule-1 and total sialic acid levels in patients with breast cancer. Eur Surg Res 2007;39:136–40.

Gogali A, Charalabopoulos K, Zampira I, et al. Soluble adhesion molecule-1 and total sialic acid levels in patients with breast cancer. Eur J Cancer 2002;38:1635–6.

Ishihara N, Matsuo H, Murakoshi H, et al. Increased apoptosis in the syncytiotrophoblast in human term placentas complicated by either preeclampsia or intrauterine growth retardation. Am J Obstet Gynecol 2002;186:158–66.

Sibai BM, Ewell M, Levine RJ, et al. Risk factors associated with either preeclampsia or intrauterine growth retardation. Am J Obstet Gynecol 2002;186:158–66.

Corson MA, James NL, Latta SE, et al. Phosphorylation of endothelial nitric oxide synthase. Am J Physiol Renal Physiol 2001;280:F93–106.

Cacanyova S. The vasoactive role of nitric oxide: physiological and morphological aspects. Curr Pharm Biotechnol 2011;12:1294–304.

Gkaliagkousi E, Ferro A. Nitric oxide signalling in the regulation of cardiovascular and platelet function. Front Biosci (Landmark Ed) 2011;16:1873–97.

Vita JA. Endothelial function. Circulation 2011;124:e906–12.

Siervo M, Coady EA, Donnelly L, et al. The role of nitric oxide oxide in endothelial function. Eur Heart J 2006;27:837–47.

Kouchoukas N, Soldin JP, Giorgi C, et al. Nitric oxide on endothelial function. Curr Vasc Pharmacol 2012;10:41–8.

van Hinsbergh VW. Endothelium – role in regulation of coagulation and inflammation. Semin Immunopathol 2012;34:93–106.

Lei J, Vodovzot Y, Tzeng E, et al. Nitric oxide, a protective molecule in the cardiovascular system. Nitric Oxide 2013;35:175–85.

Loscalzo J. The identification of nitric oxide as endothelium-derived relaxing factor. Circ Res 2013;113:100–3.

Makhlouf M, Coady EA, Donnelly L, et al. Nitric oxide synthase isozymes. Characterization, purification, molecular cloning, and functions. Hypertension 1994;23:1121–31.

Corson MA, James NL, Latta SE, et al. Phosphorylation of endothelial nitric oxide synthase in response to fluid shear stress. Circ Res 1996;79:984–91.

Ballermann BJ, Dardik A, Eng E, et al. Shear stress and the endothelium. Kidney Int Suppl 1998;67:S100–8.

Balligand JL, Feron O, Dessy C. eNOS activation by physical forces: from short-term regulation of contraction to chronic remodeling of cardiovascular tissues. Physiol Rev 2009;89:481–534.

Boon RA, Harrevoets AJ. Key transcriptional regulators of the vasoprotective effects of shear stress. Hamostaseologie 2009;29:39–40, 41–33.

Vessieres E, Freidja ML, Loufrani L, et al. Flow (shear stress)-mediated remodeling of resistance arteries in diabetes. Vascul Pharmacol 2012;57:173–8.

Govers R, Rabelink TJ. Cellular regulation of endothelial nitric oxide synthase. Am J Physiol Renal Physiol 2001;280:F193–206.

Duda DG, Fukumura D, Jain RK. Role of eNOS in neovascularization: NO for endothelial progenitor cells. Trends Mol Med 2004;10:143–5.

Sessa WC. eNOS at a glance. J Cell Sci 2004;10:143–5.

Boon RA. Redox control of endothelial function and dysfunction: molecular mechanisms and therapeutic opportunities. Antioxid Redox Signal 2008;10:1713–65.
129. Rizzo V, McIntosh DP, Oh P, et al. In situ flow activates endothelial nitric oxide synthase in luminal caveolae of endothelium with rapid caveolin dissociation and calmodulin association. J Biol Chem 1998;273:34724–9.

130. Garcia-Cardena G, Comander J, Anderson KR, et al. Biomechanical activation of vascular endothelium as a determinant of its functional phenotype. Proc Natl Acad Sci USA 2001;98:4478–85.

131. Koo A, Nordsletten D, Umerton R, et al. In silico modeling of shear-stress-induced nitric oxide production in endothelial cells through systems biology. Biophys J 2013;104:2285–306.

132. Bevilacqua MP, Pober JS, Majeau GR, et al. Interleukin 1 (IL-1) induces biosynthesis and cell surface expression of procollagen activity in human vascular endothelial cells. J Exp Med 1984;160:618–23.

133. Bevilacqua MP, Pober JS, Wheeler ME, et al. Interleukin 1 activates cultured human vascular endothelium to increase the adhesion of polymorphonuclear leukocytes, monocytes, and related leukocyte cell lines. J Clin Invest 1985;76:2003–11.

134. Yong K, Khwaja A. Leucocyte cellular adhesion molecules. Blood Rev 1990;4:211–25.

135. Bevilacqua MP, Pober JS, Mendrick DL, et al. Identification of an inducible endothelial-leukocyte adhesion molecule. Proc Natl Acad Sci USA 1987;84:9238–42.

136. Alfelda SM, Smith CW, Ward PA. Adhesion molecules and inflammatory injury. FASEB J 1994;8:504–12.

137. Jutila MA. Leukocyte traffic to sites of inflammation. APMIS 1992;100:191–201.

138. Williams TJ, Hellesweg PG. Endothelial cell biology. Adhesion molecules involved in the microvascular inflammatory response. Am Rev Resp Dis 1992;146:S45–50.

139. De Caterina R, Libby P, Peng HB, et al. Nitric oxide decreases cytokine-induced endothelial activation. Nitric oxide selectively reduces endothelial expression of adhesion molecules and proinflammatory cytokines. J Clin Invest 1995;96:60–8.

140. Liao JK. Linking endothelial dysfunction with endothelial cell activation. J Clin Invest 2013;123:540–1.

141. Grumbach IM, Chen W, Mertens SA, et al. A negative feedback mechanism involving nitric oxide and nuclear factor kappa-B modulates endothelial nitric oxide synthase transcription. J Mol Cell Cardiol 2005;39:595–603.

142. Lazzerini G, Del Turco S, Basta G, et al. Prominent role of NF-kappaB in the induction of endothelial activation by endogenous nitric oxide inhibition. Nitric Oxide 2009;21:557–62.

143. Lee KS, Kim J, Kwak SN, et al. Functional role of NF-kappaB in expression of human endothelial nitric oxide synthase. Biochem Biophys Res Commun 2014;448:101–7.

144. Panza JA, Quyyumi AA, Brush Jr. JE, et al. Abnormal endothelium-dependent vascular relaxation in patients with essential hypertension. N Engl J Med 1990;323:229:620–30.

145. Talib L, Lorin J, Zeller M, et al. Nitric oxide synthase inhibition and oxidative stress in cardiovascular diseases: possible therapeutic targets? Pharmaco Ther 2013;140:239–57.

146. Magenta A, Greco S, Capogrossi MC, et al. Nitric oxide, oxidative stress, and p66Shc interplay in diabetic endothelial dysfunction. Biomed Res Int 2014;2014:193095.
177. Ogge G, Romero R, Chaiworapongs A, et al. Leukocytes of pregnant women with small-for-gestational age neonates have a different phenotypic and metabolic activity from those of women with preeclampsia. J Matern Fetal Neonatal Med 2010;23:476–87.

178. Sacks G, Sargent I, Redman C. An innate view of human pregnancy. Immunol Today 1999;20:114–8.

179. Schiff E, Friedman SA, Baumann P, et al. Tumor necrosis factor-alpha in pregnancies associated with preeclampsia or small-for-gestational-age newborns. Am J Obstet Gynecol 1994;170:1224–9.

180. Conrad KP, Benyo DF. Placental cytokines and the pathogenesis of preeclampsia. Am J Reprod Immunol 1997;37:240–9.

181. Vitoratos N, Economou E, Iavazzo C, et al. Maternal serum levels of IL-12, interferon-gamma, SOCS3 and TGF-beta levels in pregnant women with preeclampsia, and their relation with severity of disease and fetal birth weight. J Matern Fetal Neonatal Med 2012;25:1569–73.

182. Freeman DJ, McManus F, Brown EA, et al. Short- and long-term changes in plasma inflammatory markers associated with preeclampsia. Hypertension 2004;44:708–14.

183. Jonsson Y, Ruber M, Matthiesen L, et al. Cytokine mapping of sera from women with preeclampsia and normal pregnancies. J Reprod Immunol 2006;70:83–91.

184. Sharma A, Satyam A, Sharma JB. Leptin, IL-10 and inflammatory markers (TNF-alpha, IL-6 and IL-8) in pre-eclamptic, normotensive pregnant women. Mediators Inflamm 2010;2010:598649.

185. Borecki B, Aksoy H, Al RA, et al. Maternal serum interleukin-10, interleukin-2 and interleukin-6 in pre-eclampsia and eclampsia. Am J Reprod Immunol 2007;58:56–64.

186. Gu Y, Lewis DF, Deere K, et al. Elevated maternal IL-16 levels, enhanced IL-16 expressions in endothelium and leukocytes, and increased IL-16 production by placental trophoblasts in women with preeclampsia. J Immunol 2008;181:4418–22.

187. Sibai B, Romero R, Klebanoff MA, et al. Maternal plasma concentrations of the soluble tumor necrosis factor receptor 2 are increased prior to the diagnosis of preeclampsia. Am J Obstet Gynecol 2009;200:630.e1–8.

188. Lewis DF, Canzoneri BJ, Wang Y. Maternal circulating TNF-alpha levels are highly correlated with IL-10 levels, but not IL-6 and IL-8 levels, in women with pre-eclampsia. Am J Reprod Immunol 2009;62:269–74.

189. Vitoratos N, Economou E, Iavazzo C, et al. Serum levels of TNF-alpha and IL-6 long after delivery in preeclamptic and normotensive pregnant women. Mediators Inflamm 2010;2010:98649.

190. Tosun M, Celik H, Avci B, et al. Maternal and umbilical serum levels of interleukin-6, interleukin-8, and tumor necrosis factor-alpha in normal pregnancies and in pregnancies complicated by preeclampsia. J Matern Fetal Neonatal Med 2010;23:880–6.

191. Kalkunte S, Nevers T, Norris WE, et al. Vascular IL-10: a protective role in preeclampsia. J Reprod Immunol 2011;88:165–9.

192. Xiao JP, Yin XY, Gao YF, et al. The increased maternal serum levels of IL-6 are associated with the severity and onset of preeclampsia. Cytokine 2012;60:856–60.

193. Cemgil Arkan D, Aral M, Coskun A, et al. Plasma IL-4, IL-8, IL-12, interferon-gamma and CRP levels in pregnant women with preeclampsia, and their relation with severity of disease and fetal birth weight. J Matern Fetal Neonatal Med 2012;25:1569–73.

194. Lau SY, Guild SJ, Barrett CJ, et al. Tumor necrosis factor-alpha, interleukin-6, and interleukin-10 levels are altered in preeclampsia: a systematic review and meta-analysis. Am J Reprod Immunol 2013;70:412–27.

195. Ozkan ZS, Simsek M, Ilhan F, et al. Plasma IL-17, IL-35, interferon-gamma, SOCS3 and TGF-beta levels in pregnant women with preeclampsia, and their relation with severity of disease. J Matern Fetal Neonatal Med 2014;27:1513–7.

196. Pinheiro MB, Martins-Filho OA, Mota AP, et al. Severe preeclampsia goes along with a cytokine network disturbance towards a systemic inflammatory state. Cytokine 2013;62:165–73.

197. Sahin S, Ozakpinar OB, Ergolu M, et al. The impact of platelet functions and inflammatory status on the severity of preeclampsia. J Matern Fetal Neonatal Med 2014;26:1–6.

198. Mellembakken JR, Aukrust P, Hestad K, et al. Chemokines and leukocyte activation in the fetal circulation during preeclampsia. Hypertension 2001;38:394–8.

199. Gotsch F, Romero R, Friel L, et al. CXCL10/IP-10: a missing link between inflammation and angiogenesis in preeclampsia? J Matern Fetal Neonatal Med 2007;20:777–92.

200. Laskowska M, Laskowska K, Leszczynska-Gorzela B, et al. Comparative analysis of the maternal and umbilical interleukin-8 levels in normal pregnancies and in pregnancies complicated by preeclampsia with intrauterine normal growth and intrauterine growth retardation. J Matern Fetal Neonatal Med 2007;20:527–32.

201. Boj R, Svensson J, Nilsson-Ekdahl K, et al. Biomarkers of coagulation, inflammation, and angiogenesis are independently associated with preeclampsia. Am J Reprod Immunol 2012;68:258–70.

202. Molvarec A, Szarka A, Walentin S, et al. Serum leptin levels in relation to circulating cytokines, chemokines, adhesion molecules and angiogenic factors in normal pregnancy and preeclampsia. Reprod Biol Endocrinol 2011;9:124.

203. Da M, Basu A, Fu D, et al. Serum inflammatory markers and preeclampsia in type 1 diabetes: a prospective study. Diabetes Care 2013;36:2054–61.

204. Guven MA, Coskun A, Ertas IE, et al. Association of maternal serum CRP, IL-6, TNF-alpha, homocysteine, folic acid and vitamin B12 levels with the severity of preeclampsia and fetal birth weight. Hypertens Pregnancy 2009;28:190–200.

205. Stampalija T, Chaiworapongsa T, Romero R, et al. Maternal plasma concentrations of sST2 and angiogenic/anti-angiogenic factors in preeclampsia. J Matern Fetal Neonatal Med 2013;26:1359–70.

206. Granne I, Southcombe JH, Snider JV, et al. ST2 and IL-33 in pregnancy and pre-eclampsia. PLoS One 2011;6:e24463.

207. Wang Y, Lewis DF, Gu Y, et al. Elevated maternal soluble gp130 and IL-6 levels and reduced gp130 and SOCS-3 expressions in women complicated with preeclampsia. Hypertension 2011;57:336–42.

208. Sharma D, Singh A, Trivedi SS, et al. Role of endothelin and inflammatory cytokines in pre-eclampsia – a pilot North Indian study. Am J Reprod Immunol 2011;65:428–32.

209. Ozler A, Turgut A, Sak ME, et al. Serum levels of neopterin, tumor necrosis factor-alpha and Interleukin-6 in preeclampsia: relationship with disease severity. Eur Rev Med Pharmacol Sci 2012;16:1707–12.

210. Mosimann B, Wagner M, Poon LC, et al. Maternal serum cytokines at 30–33 weeks in the prediction of preeclampsia. Prenat Diagn 2013;33:823–30.

211. Zhang Z, Gao Y, Zhang L, et al. Alterations of IL-6, IL-6R and gp130 in early and late onset severe preeclampsia. Hypertens Pregnancy 2013;32:270–80.

212. Darling AM, McDonald CR, Conroy AL, et al. Angiogenic and inflammatory biomarkers in midpregnancy and small-for-gestational-age outcomes in Tanzania. Am J Obstet Gynecol 2014. [Epub ahead of print].

213. Visser S, Hermes W, Ket JC, et al. Systematic review and metaanalysis on nonclassic cardiovascular biomarkers after hypertension associated with small-for-gestational-age outcomes in Tanzania. Am J Obstet Gynecol 2014. [Epub ahead of print].

214. Tjoa ML, van Vught JM, Go AT, et al. Elevated C-reactive protein levels during first trimester of pregnancy are indicative of preeclampsia and intrauterine growth restriction. J Reprod Immunol 2003;59:29–37.

215. Qiu C, Luthy DA, Zhang C, et al. A prospective study of maternal serum C-reactive protein concentrations and risk of preeclampsia. Am J Hypertens 2004;17:154–60.

216. Teran E, Escudero C, Calle A. C-reactive protein during normal pregnancy and preeclampsia. J Int J Gynaecol Obstet 2005;93:299–300.

217. Paternoster DM, Fantinato S, Stella A, et al. C-reactive protein in hypertensive disorders in pregnancy. Clin Appl Thromb Hemost 2006;12:330–7.

218. Garcia RG, Celedon J, Sierra-Laguardo J, et al. Raised C-reactive protein and impaired flow-mediated vasodilation precede the development of preeclampsia. Am J Hypertens 2007;20:98–103.
219. Ertas IE, Kahyaoglu S, Yilmaz B, et al. Association of maternal serum high sensitive C-reactive protein level with body mass index and severity of pre-eclampsia at third trimester. J Obstet Gynaecol Res 2010;36:970–7.

220. de Jonge LL, Steegers EA, Ernst GD, et al. C-reactive protein levels, blood pressure and the risks of gestational hypertensive complications: the Generation R Study. J Hypertens 2011;29:2413–21.

221. Gandevani SB, Banaem LM, Mohamadi B, et al. Association of high-sensitivity C-reactive protein serum levels in early pregnancy with the severity of preeclampsia and fetal birth weight. J Perinat Med 2012;14:1–5.

222. Kucukgoz Gulec U, Tuncay Ozgunen F, Baris Guzel A, et al. An analysis of C-reactive protein, procalcitonin, and D-dimer in pre-eclamptic patients. Am J Reprod Immunol 2012:68:331–7.

223. Chaemsaithong P, Chaiworapongsa T, Romero R, et al. Fetuin-A, a negative acute phase protein reactant, is decreased in preeclampsia. J Matern Fetal Neonatal Med 2014;27:217–27.

224. Rebelo F, Schlussel MM, Vaz JS, et al. C-reactive protein and later complications: systematic review and meta-analysis taking into account the weight status. J Hypertens 2013;31:16–26.

225. Chaemsaithong P, Romero R, Chaiworapongsa T, et al. Maternal plasma soluble TRAIL is decreased in preeclampsia. J Matern Fetal Neonatal Med 2014;27:167–78.

226. Waage A, Aasen AO. Different role of cytokine mediators in septic shock related to meningococcal disease and surgery/polytrauma. Immunol Rev 1992;127:221–30.

227. Martin C, Boisson C, Haccoun M, et al. Patterns of cytokine evolution (tumor necrosis factor-alpha and interleukin-6) after septic shock, hemorrhagic shock, and severe trauma. Crit Care Med 1997;25:1813–9.

228. Hack CE, Aarden LA, Thijs LG. Role of cytokines in sepsis. Adv Immunol 1997;66:101–95.

229. Riche FC, Cholley BP, Panis YH, et al. Inflammatory cytokine response in patients with septic shock secondary to generalized peritonitis. Crit Care Med 2000;28:433–7.

230. Romero R, Kadar N, Vaisbuch E, et al. Maternal death following antepartum peritonitis. Crit Care Med 2000;28:433–7.

231. Mikkelsen ME, Shah CV, Scherpereel A, et al. Lower serum endocan levels are associated with the development of acute lung injury after major trauma. J Crit Care 2012;27:522.e11–7.

232. Cunningham FG, Leveno KJ, Hankins GD, et al. Respiratory insufficiency associated with preeclampsia during pregnancy. Obstet Gynecol 1984;63:121–5.

233. Cunningham FG, Lucas MJ. Urinary tract infections complicating pregnancy. Baillieres Clin Obstet Gynaecol 1994;8:353–73.

234. Cunningham FG, Lucas MJ. Pulmonary injury complicating antepartum preeclampsia. Am J Obstet Gynecol 1987;156:797–807.

235. Jolley JA, Kim S, Wing DA. Acute pyelonephritis in pregnancy: an 18-year retrospective analysis. Am J Obstet Gynecol 2014;210:219.e1–6.