

The Relation Between von Willebrand Factor Plasma Concentration and Ultrasonographic Doppler Findings in Pregnancies Complicated by Hypertensive Disorders

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Abstract

Background: Hypertensive disorder of pregnancy is one of the most common complications in pregnancy forming a triad together with hemorrhage and infection. It affects about 10% of pregnancies and contributes for a significant maternal and perinatal mortality.

Aim: The aim of this study was to evaluate the relationship between maternal plasma VWF level and fetal Doppler flow measurements in pregnancies complicated by hypertensive disorders

Patients and methods: A case-control study was conducted in Tikrit city from March to September 2019. From a total of 100 enrolled pregnant women, 65 were eligible for the final analyses. 35 were excluded because they did not meet the inclusion criteria. The 65 women were placed into the following groups: pregnant women with chronic hypertension ($n = 25$), those with gestational hypertension ($n = 20$), and those with preeclampsia ($n = 20$). These pregnant women were included in the study when attended Salah Al-Din General Hospital, Labor room. A control group of 22 healthy normotensive pregnant females was also included. The Doppler flow measurements of uterine arteries, umbilical artery, and middle cerebral artery were obtained by ultrasound (Philips 6Hd 11EX) using a convex probe (frequency 3.5–5 MHz). Measurements were taken when the fetus was not moving and did not have any breathing movements. Venous blood was collected for to measurement vWf by ELISA

Results: The study demonstrated that the highest mean of VWF was observed in pregnant women with Preeclampsia (5.71 ± 1.14 pg/ml) followed by gestational hypertension group (4.33 ± 0.60 pg/ml) and chronic hypertension group (3.29 ± 0.55 pg/ml) and the lowest mean was within the control group (1.66 ± 0.41 pg/ml) respectively (P. value < 0.01). The study found increased vWf plasma concentrations in preeclamptic patients was associated with abnormal Doppler finding with no significant relation between gestational age of women with preeclamptic and abnormal Doppler finding

Conclusions: Increased vWf plasma concentration corresponds to ultrasonographic features of placental insufficiency in women with preeclampsia. This relationship may confirm severity of endothelial damage in preeclamptic patients. Widely available and relatively low-cost Doppler flow measurements may be useful for selecting women at higher risk of developing cardiovascular diseases.

Keywords: Von Willebrand; Doppler Findings; Pregnancy; Hypertensive Disorders; Preeclampsia

Introduction

Hypertensive disorder of pregnancy is one of the most common complications in pregnancy forming a triad together with hemorrhage and infection. It affects about 10% of pregnancies and contributes for a significant maternal and perinatal mortality⁽¹⁾. The World Health Organization (WHO) reported that 14.0% of global maternal deaths are attributed to hypertensive disorders of pregnancy⁽²⁾. Hypertension in pregnancy is defined as a systolic of 140 mm Hg or greater or a diastolic of 90 mm Hg or greater. Blood pressure should be taken in the upper arm with the patient seated using an appropriately sized cuff⁽³⁾. Hypertension in pregnancy is said to occur when the systolic blood pressure is ≥ 140 mmHg and diastolic blood pressure is ≥ 90 mmHg on at least two occasions, six hours apart. Hypertensive disorder of pregnancy encompasses gestational hypertension in pregnancy (hypertension without proteinuria), pre-eclampsia (hypertension with proteinuria) and eclampsia (pre-eclampsia with convulsions)⁽⁴⁾. Studies have attempted to explain the pathophysiology of hypertension in pregnancy; however, a consensus is lacking^(5,6). Recent evidence suggests that impaired placental vascularization is a major factor responsible for gestational

hypertension development; however, the etiology is probably multifactorial. It has been suggested that impaired cytotrophoblast proliferation and migration are present, as is cell differentiation to endothelial cells in uterine arteries, and as a result there is impaired trophoblast invasion to placental blood vessels^(7,8). Scientists agree about the 2-stage model of preeclampsia development. The first stage begins just after trophoblast-impaired implantation that causes hypoxia. In this stage, impaired uterine artery may be observed in Doppler flows⁽⁹⁾. The second stage, when patients typically develop clinical symptoms, begins mostly after the 20th week of gestation and is caused by placental factors released into maternal blood⁽⁵⁰⁾. As a result, systemic inflammation begins to damage endothelium in many organs, which is followed by typical preeclampsia symptoms⁽¹⁰⁾. However, the potential mechanisms of these changes have not been decisively explored. Von Willebrand factor (VWF) connects platelets with endothelial cells, collagens, and is responsible for the correct platelet-adhesion process^(11,12). The aim of this study to evaluate the relationship between maternal plasma VWF level and fetal Doppler flow measurements in pregnancies complicated by hypertensive disorders

Patients and methods

A case-control study was conducted in Tikrit city in Salah Al din general hospital from first of March 2019 and to end of September 2019. From a total of 100 enrolled pregnant women, 65 were eligible for the final analyses. 35 were excluded because they did not meet the inclusion criteria. The 65 women were placed into the following groups: pregnant women with chronic hypertension ($n = 25$), those with gestational hypertension ($n = 20$), and those with preeclampsia ($n = 20$), patients were defined and divided according to the National High Blood Pressure Education Program Working Group on High Blood Pressure in Pregnancy. A control group of 22 healthy normotensive pregnant females was also included. All comparison groups were matched for mother's age, parity, and gestational age at the time of enrollment and blood collection.

Inclusion criteria

1. Singleton pregnant women with chronic hypertension.
2. Singleton pregnant with gestational hypertension.
3. Singleton pregnant with preeclampsia.
4. Age: 15-45 year
5. Parity: 1-7.

Exclusion criteria

1. Multiple gestations.
2. Fetal structural/genetic anomalies.
3. Maternal renal disease.
4. Metabolic disease.
5. Inflammatory disease.
6. Autoimmune disease, and other comorbidities associated with endothelial damage.

Methods

Hypertensive disorders were defined according to classification of The National High Blood pressure Education program Working group on High Blood pressure in pregnancy. Twenty-four-hour automatic blood pressure monitoring was performed Blood pressure readings were taken at 30minute intervals. For patients who was admitted to the hospital ,and two readings of blood pressure was taken for patients who was not admitted to the hospital and diagnosed as hypertensive disorders depend on history and examination and blood pressure readings that record on antenatal card in each antenatal visit. Measurement of Bp done by sphygmomanometer in sitting position, with cuff size appropriate to patients arm circumference was used, and to eliminate possible stressor for the patients, the visual preview of blood pressure measurement was removed.

Three ml of blood was collected by vein puncture, blood samples were placed into sterile test tubes and left for 30 minutes at 37 °C then were centrifuged at 3000 rpm for 15 minutes then the clot was removed and the obtained sera were then aspirated using automatic micropipette and transferred into clean test tubes and stored in deep freeze at -20°C for determination of vWF by using ELISA technique

Every woman had ultrasound examination with Doppler flow measurements of uterine arteries, umbilical vessels, and fetal middle cerebral artery, simultaneously with recruitment into study and blood sample collection. Doppler done by 2 radiologists and they confirmed the same results. The Doppler flow measurements of uterine arteries, umbilical artery, and middle cerebral artery were obtained by ultrasound (Philips 6Hd 11EX) using a convex probe (frequency 3.5–5 MHz). Measurements were taken when the fetus was not moving and did not have any breathing movements.

Results

As shown in Table 1. There was no significant difference between studied cases and the control group regarding patient age and gestational age at sampling and parity ($P > 0.05$) while there was a significant difference between studied cases and the control group regarding systolic and diastolic blood pressure ($P < 0.05$).

Table 1: Clinical characteristics of studied women

Parameters (Mean±SD)	Chronic hypertension	Gestational hypertension	Pre-eclampsia	Control group
No.	25	20	20	22
Maternal age (yeas)	32.5±4.2	30.1±4.1	32.2±5.9	32.2±6.2
Gestational age	33.4±7.5	36.8±4.1	34.1±3.2	35.4±6.6
Parity, median (Range)	2 (1–7)	2 (1–4)	1 (1–6)	2 (1–6)
Mean 24 h SBP, mm Hg	128.7±11.5 *	133.4±13.1*	148.6±14.3*	110.8±7.4
Mean 24 h DBP, mm Hg	83.5±6.9*	84.7±9.9*	96.6±12.9*	69.4±8.3
Maximal SBP, mm Hg	167±24.1*	153.9±18.2*	177.9±24.1*	119.0±10.2
Maximal DBP, mm Hg	109.1±22.7*	105.8±17.0*	118.8±12.8*	74.7±13.3

* Statistically significant differences ($p < 0.05$) between studies and control group.

SBP, systolic blood pressure; DBP, diastolic blood pressure.

No statistically significant differences existed between groups in (T. Test $p = NS$).

The study demonstrated that VWF mean was significantly elevated in pregnant women with chronic hypertension as compared with the control group (3.29 ± 0.55 and 1.66 ± 0.41 pg/ml) respectively (P . value < 0.01)

The study demonstrated that the highest mean of VWF was observed in pregnant women with Pre-eclampsia followed by gestational hypertension group and chronic hypertension group and the lowest mean was within the control group (P . value < 0.001), as shown Table 3.

Table 3: Comparison of the von Willebrand factor among the studied groups.

VWF (pg/ml)	Chronic hypertension	Gestational hypertension	Pre-eclampsia	Control group	ANOVA	P. value
N	25	20	20	22	118.4	0.0001
Mean	3.29	4.33	5.71	1.66		
SD.	0.55	0.60	1.14	0.41		

In Table 4, the results are provided for Doppler flow measurements in study and control groups.

Table 4: Doppler ultrasound results in each patient group

	Chronic hypertension	Gestational hypertension	Pre-eclampsia	Control group
PI-UA	1.2±0.27*	1.09±0.28	1.33±0.43*	0.71±0.17

(mean ± SD)				
PI-MCA (mean ± SD)	1.59±0.43	1.59±0.33	1.42±0.39*	1.81±0.51
CUR (mean ± SD)	1.49±0.54*	1.61±0.71*	1.29±0.55*	2.31±0.55
PI-RtUtA (mean ± SD)	1.11±0.5	1.21±0.83*	1.35±0.43*	0.44±0.21
PI-LtUtA (mean ± SD)	2.16±1.22*	0.91±0.55	1.71±0.65*	0.74±0.17
UAS (median; minimum – maximum)	2 (0–4)	0 (0–4)	3 (0–4)	0 (0–0)

* Statistically significant differences compared with those of the control group ($p < 0.05$; Mann-Whitney test). PI-UA, umbilical artery pulsatility index; PI-MCA, middle cerebral artery pulsatility index; CUR, cerebroumbilical ratio; PI-RtUtA, right uterine artery pulsatility index; PI-LtUtA, left uterine artery pulsatility index; UAS, uterine artery score.

3.4. Relation of von Willebrand factor and Doppler finding in women with preeclampsia

The study found increased vWf plasma concentrations in preeclamptic patients was associated with abnormal Doppler finding (Table 5).

Table 3.7: Relation of von Willebrand factor and Doppler finding in women with preeclampsia

VWF (pg/ml)	Normal Doppler finding	Abnormal Doppler finding
N	7	13
Mean	3.96	6.41
SD.	0.59	1.21

P. value: 0.001

Discussion

The study demonstrated that the highest mean of VWF was observed in pregnant women with Preeclampsia followed by gestational hypertension group and chronic hypertension group and the lowest mean was within the control group (P. value <0.001), as shown Table 3. More recent studies have confirmed that in hypertension, vWf deposition in placental vessel walls may lead to their hyperplasia, impaired endothelial regeneration, and limited angiogenesis^(7,8,9). All these changes may lead to increased vessel resistance in Doppler ultrasound measurements. Soares *et al*⁽¹⁰⁾ evaluated women with type 2 diabetes using ultrasonography Doppler for assessment of carotid intima-media thickness. Elevated vWf plasma concentration was found only in patients with moderate macrovascular disease (≥ 1 mm and presence of plaque and stenosis) but was not detected in women without or with mild macrovascular disease. Molvarec *et al*⁽¹¹⁾ confirmed a correlation between increased vWf and soluble fms-like tyrosine kinase-1 (sFlt-1) plasma concentration in preeclamptic patients. Another study revealed that in late-onset preeclampsia, an imbalance existed in circulating angiogenic and anti-angiogenic factors already present at 8–10 weeks of pregnancy. Decreased PlGF plasma concentration, increased sFlt-1 plasma concentration, and increased sFlt-1/PlGF ratio were observed in these patients. Moreover, abnormal first trimester uterine arteries were found on ultrasound Doppler, and placental insufficiency was confirmed in histopathology after delivery⁽¹²⁾. It is highly probable that sFlt-1 and vWf play a key role in the abnormal placentation, which is followed by symptoms of preeclampsia. It is well known that higher plasma concentrations of vWf are associated with hypertension and progression of cardiovascular disease⁽¹³⁾, including stroke, myocardial infarction, and venous thrombosis⁽¹⁴⁾. Furthermore, infactors for the development of microvascular complications in patients with diabetes mellitus⁽¹⁵⁾. The multimeric vWf is proteolytically regulated by the ADAMTS13 (a disintegrin and metalloproteinase with a thrombospondin type 1 motif, member 13). The ADAMTS13 is a metalloprotease enzyme that splits large, thrombogenic vWf multimers, decreasing their activity⁽¹⁶⁾. The

importance of ADAMTS13 in supporting the balance of vWf multimeric size is well presented by its role in some hematologic diseases, for instance, idiopathic thrombotic thrombocytopenic purpura and some cases of von Willebrand disease. Many reports in the current literature demonstrate the association between high vWf plasma concentrations and/or low ADAMTS13 plasma levels with increased risk of thrombotic diseases⁽¹⁷⁻²²⁾. Based on these studies, vWf may be a promising target in the prevention and eventually treatment of thrombotic diseases, and recombinant ADAMTS13 could be a novel therapeutic agent for use in such disorders⁽²³⁾. Furthermore, some experimental animal models of ischemic stroke and myocardial infarction supported vWf as a potential therapeutic target⁽²⁴⁾. The Doppler flow measurements for evaluating placental efficiency and eventually the assessment of vWf plasma concentration in preeclamptic women may identify the patients at much higher risk of cardiovascular diseases in the future and may qualify them for such prevention and eventually therapy. vWf is synthesized by endothelial cells and megakaryocytes. Significantly higher plasma vWf concentrations have been found in pregnancies complicated by gestational hypertension and preeclampsia compared with healthy pregnancies⁽⁸⁾. Several studies have found a correlation between abnormal uterine artery Doppler flow and preeclampsia development. In 2008, Brodzki *et al*⁽²⁷⁾ reported that bilateral notch sign in uterine arteries and elevated resistance in maternal-fetal circulation corresponds to more advanced endothelial damage. Moreover, it has conclusively been shown that increased vascular resistance in the course of preeclampsia is followed by abnormal middle cerebral and umbilical artery flow, especially in intrauterine growth-restricted fetuses⁽¹¹⁾.

Conclusions

- 1- The highest mean of VWF was observed in pregnant women with Pre-eclampsia followed by gestational hypertension group and chronic hypertension group and the lowest mean was within the control group
- 2- This relationship may confirm severity of endothelial damage in preeclamptic patients.
- 3- Widely available and relatively low-cost Doppler flow measurements may be useful for selecting women at higher risk of developing cardiovascular diseases.

Recommendations

- 1- Future investigations need with large population studies regarding the relation of vWf with hypertension pregnancy.
- 1- Further studies required for evaluate the relation of vWf level with pre-eclampsia in first months of pregnancy.

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