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FULL PAPER

The differences of serum levels of sFlt-1, PlGF, and sFlt-1/PlGF ratio throughout daytime and nighttime in women with severe preeclampsia at 30-34 weeks of pregnancy: a pilot study

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Preeclampsia is identifiable by an imbalance of angiogenic factors, which entails elevated levels of sFlt-1 and a reduction of PlGF levels. In addition to disruptions in circadian rhythms, preeclamptic individuals may exhibit daytime and nighttime alterations in sFlt-1, PlGF, and the sFlt-1/PlGF ratio. This study aims to assess the differences in daytime and nighttime concentrations of sFlt-1, PlGF, and the sFlt-1/PlGF ratio in severe preeclamptic women between 30 and 34 weeks of gestational age. This cross-sectional study was conducted at Dr. Soetomo Public Academic Hospital and Airlangga University Hospital in Surabaya, involving ten pregnant women at 30-34 weeks of gestation, all diagnosed with severe preeclampsia. Samples of blood were drawn in the morning at 08:00, followed by the evening at 20:00. The study found no significant differences in serum levels of sFlt-1 (5.99 ± 3.29 vs. 6.82 ± 4.09 pg/mL), PlGF (85.5 [37-312] vs. 72 [35-437] pg/mL), and the sFlt-1/PlGF ratio (48.5 [6-233] vs. 58.5 [3-345] pg/mL) between day and night (p > 0.05). The results suggest that the circadian rhythm does not notably influence the serum levels of anti- and proangiogenic substances in confirmed severe preeclampsia.

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Introduction

Preeclampsia is hypertension that develops at or after the twentieth week of pregnancy, along with urine protein, organ failure, and uteroplacental impairment. This condition ranks as one of the most significant global problems involving the deaths of pregnant women. It causes approximately 76,000 fatal maternal deaths and 500,000 deaths of newborns annually [1-4]. Preeclampsia is a "disease of theories". The aberrant placentation hypothesis is the most accepted currently. The stress of syncytiotrophoblast and endothelial tissue cell reactivity are the core features of the pathognomonic process, significantly impacting the angiogenic substances involving Placental Growth Factor (PlGF) and Soluble Fms-like Tyrosine Kinase 1 (sFlt-1), leading to an angiogenic imbalance

and subsequent manifestation of the clinical hallmarks of preeclampsia [5-9].

PlGF represents a proangiogenic factor, while sFlt-1 represents an antiangiogenic factor. In preeclamptic patients, PlGF levels decrease, and sFlt-1 levels increase, resulting in an elevated sFlt-1/PlGF ratio [10-14]. Several countries extensively utilize this biomarker for the prediction, diagnosis, and management of preeclampsia [12,13,15,16].

Antiangiogenic and proangiogenic levels and ratios might fluctuate day and night. Cortisol and melatonin could impact alterations in biomarkers. Circadian rhythms physiologically influence these hormones [17-21]. Preeclamptic patients could disrupt those hormone releases due to circadian rhythm abnormalities [22-24].

This study suggests alterations throughout the day and night in anti- and proangiogenic factors and their ratios in severe preeclamptic patients. Previous studies randomly obtained blood samples for these biomarkers at various times of the day, even over weeks, without emphasizing a specific sampling time [25,26]. No study has investigated the daytime and nighttime alterations and characteristics of sFlt-1, PlGF, and the ratio of them in severe preeclamptic pregnancy nationwide. Thus, this pilot study is essential to determine day-to-night fluctuations in those angiogenic factors in confirmed severe preeclampsia at 30-34 weeks gestation.

Methods

This cross-sectional study was conducted at the tertiary hospitals Dr. Soetomo Public Academic Hospital and Airlangga University Hospital in Surabaya. It involved ten pregnant women at 30 to 34 weeks of gestation who met the specified inclusion and exclusion criteria. The inclusion criteria encompassed singleton pregnant women diagnosed with severe preeclampsia between thirty to thirty-four weeks of gestation, receiving expectant management. In contrast, the exclusion criteria included labor conditions, placenta accreta spectrum, malignancy during pregnancy, gestational and pregestational diabetes mellitus, and superimposed chronic hypertension. These women were diagnosed with severe preeclampsia based on the criteria set forth by the International Society for the Study of Hypertension in Pregnancy (ISSHP) [3].

Participants were selected using a sequential sampling method. To determine the level in the serum of sFlt-1, PlGF, and their ratio, venous blood specimens were subsequently drawn at 8:00 and 20:00. The chosen timing, aligned with the circadian rhythm, was intended to capture potential fluctuations in cortisol and melatonin hormone levels. While this study did not measure confounding factors such as anxiety during pregnancy, it is acknowledged that these factors might impact hormone levels [18,27]. The analysis of angiogenic factors utilized Roche Elecsys®. The Elecsys reagent kit and COBAS e601 analyzer's Electrochemiluminescence Immunoassay (ECLIA) were utilized to calculate the serum sFlt-1 and PlGF levels. The Elecsys PlGF ranges from 3 to 10,000 pg/mL, with levels below three categorized as < 3 and excess values as > 10,000. Elecsys sFlt-1 ranges from 10 to 85,000 pg/mL, with levels less than ten classified as < 10 and values higher than eighty-five thousand as > 85,000 [28,29].

The assessment of day-and-night serum anti- and proangiogenic factors commenced with a normality test using the Shapiro-Wilk test. The hypotheses were validated with a paired t-test. Version 25 of SPSS was used for the analysis of statistical data. For normally distributed data, the paired t-test was utilized, while the Wilcoxon signed-rank test was employed for data with abnormal distributions. The significance level for this pilot study was set at 0.05, indicating significance if $p < 0.05$ from the statistical test.

Results

A total of ten patients participated in this pilot study, with six at Dr. Soetomo Public Academic Hospital and four at Airlangga University Hospital. The average body mass index of the patients recruited for this study was 33.71 kg/m², while their mean age was 32.9 years (Table 1).

According to this study, the average expectant woman diagnosed with severe preeclampsia is obese. In addition, 50% of patients in this study were primiparous, and the other 50% were multiparous. The median gestational age of patients presenting for the first time was 31 weeks. An analysis of variance was conducted on the daytime and nighttime serum levels of sFlt-1, PlGF, and their ratio among severe preeclamptic pregnancies between thirty and thirty-four weeks of gestation.

TABLE 1 Sample characteristics

Characteristics	Total n = 10 (100%)	Mean ± SD/ Median (min-max)
Age (Years)		32.9 ± 6.15
Weight (kg)		79.00 ± 16.9
Height (cm)		152.9 ± 6.2
Body Mass Index (kg/m²)		33.71 ± 6.39
Duration of Marriage (Years)*		2 (0.66–15)
Initial Admission Systolic Blood Pressure (mmHg)*		164 (161–190)
Initial Admision Diastolic Blood Pressure (mmHg)		105 ± 9.45
Initial Admission Gestational Age (Weeks)*		31 (30-33)
Parity		
Nulliparous	5 (50%)	
Multiparous	5 (50%)	

The average serum sFlt-1 concentration was 5,992 ± 3,291 pg/mL during the day and 6,817 ± 4,088 pg/mL at night. Table 2 revealed that both comparable periods were not significantly different (p = 0.063).

TABLE 2 Differences in daytime and nighttime serum levels of sFlt-1

Parameter	Mean ± SD	p-value
Daytime sFlt-1 serum (pg/mL)	5,992 ± 3,291	0.063
Nighttime sFlt-1 serum (pg/mL)	6,817 ± 4,088	

sFlt-1: Soluble Fms-like Tyrosine Kinase 1

The median morning serum PlGF level was 85.5 pg/mL, while the nighttime serum PlGF level was 72.0 pg/mL. Table 3 illustrates an insignificant difference in serum PlGF values throughout the two phases of the day (p = 0.445).

TABLE 3 Differences in daytime and nighttime serum levels of PlGF

Parameter	Median (min-max)	p-value
Daytime PlGF serum (pg/mL)	85.5 (37–312)	0.445
Nighttime PlGF serum (pg/mL)	72.0 (35–437)	

PlGF: *Placental Growth Factor*

The median value for the morning sFlt-1/PlGF ratio group was 48.5, while it was 58.5 for the night group. There was no significant difference between the daytime and nighttime ratios of sFlt-1/PlGF (p = 0.241), as depicted in Table 4.

TABLE 4 Differences in the ratio of sFlt-1/PlGF in daytime and nighttime

Parameter	Median (min-max)	*P*-value
Daytime sFlt-1/PlGF Ratio	48.5 (6–233)	0.241
Nighttime sFlt-1/PlGF Ratio	58.5 (3–345)	

sFlt-1: *Soluble Fms-like Tyrosine Kinase 1*, PlGF: *Placental Growth Factor*

Discussion

The preliminary investigation attempted to evaluate the fluctuation in sFlt-1 and PlGF levels throughout the day and night in severe preeclampsia. However, the serum concentrations of proangiogenic and antiangiogenic substances did not differ significantly between the periods of the day and night. This outcome might occur due to the confounding factors influencing the concentration of these biomarkers.

Theoretically, in a normal pregnancy, blood pressure displays a diurnal rhythm, exhibiting higher daytime than nighttime levels, akin to non-pregnant patterns. Nighttime typically sees a 10-20% reduction in blood pressure, termed "normal dipping." Conversely, women with preeclampsia often experience "reverse dipping," where blood pressure exceeds daytime levels due to disrupted circadian rhythms. Reports have highlighted disruptions in placental circadian rhythms in preeclamptic women [9,23,30-32]. Maternal hormonal abnormalities may disrupt circadian rhythms and contribute to pregnancy complications. Cortisol and melatonin are the primary influences on circadian rhythms and are vital to this mechanism [22,24,33-35].

The adrenal gland releases cortisol in a circadian rhythm, resulting in elevated morning cortisol levels and a gradual decline in nocturnal levels. Disruptions in cortisol secretion lead to fluctuations in daytime and nighttime exposure [24,36-38]. The pineal gland is responsible for the production of lipophilic melatonin (5-Methoxy-N-Acetyltryptamine). However, the placenta produces a great deal of it during pregnancy. The placenta produces more melatonin than the pineal gland- the normal melatonin levels during pregnancy peak at night and decline during the day. Abnormalities in melatonin secretion influence the maternal circulation of melatonin [23,33,39-41]. These circumstances might influence the placental synthesis of angiogenic factors, potentially altering their serum levels and ratios throughout the day and night. This phenomenon, driven by disrupted circadian rhythms in severe preeclampsia, results in a reverse dipping effect, causes an increase in blood pressure during the night compared to daytime levels [22-23,31,42]. Hypertension, a manifestation of preeclampsia due to endothelial dysfunction resulting from an imbalance in angiogenic factors, tends to worsen at night. This exacerbation may lead to elevated sFlt-1 and PlGF levels at night, consequently impacting the increased sFlt-1/PlGF ratio. Elevated nocturnal blood pressure could potentially lead to kidney and cardiac damage [9,17,43-45]. This study reveals a gap in the analysis of antiangiogenic and proangiogenic factors and their ratio between day and night, but no differences in sFlt-1, PlGF, and the ratio of them in severe preeclamptic patients using statistical analysis. Supposedly, this may occur due to confounding factors. Previous studies collected blood samples for this biomarker randomly throughout the day and even weekly, lacking specific guidelines for the precise timing of biomarker measurements [25,26].

This study demonstrated that biomarker testing might be performed at any time during the day. However, it emphasizes the necessity of more extensive research to determine the most suitable timing for obtaining biomarker blood samples to ensure accuracy. Furthermore, the relationship between

circadian rhythms and anti- and proangiogenic factors demands further research. Several limitations are evident in this study. First, the study included only ten participants from two specific tertiary hospitals, limiting the generalizability of the findings to a broader population. The small sample size may not adequately represent the diverse characteristics of severe preeclampsia patients. Second, the study assumed a consistent circadian rhythm impact on biomarker levels without accounting for potential variations in individual circadian patterns among participants.

Conclusion

Serum concentrations of sFlt-1, PlGF, and their ratio did not fluctuate drastically day and night among patients diagnosed with severe preeclampsia. The angiogenic factors could be examined at any time of the day. The precise timing for collecting a blood sample for this biomarker should be investigated in greater depth. In addition, the effect of cortisol and melatonin substances on diurnal and nocturnal fluctuations in antiangiogenic and proangiogenic biomarkers requires further investigation.

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Authors' contributions

All authors (D.W.P. and M.I.A.A.) contributed in some manner to this research, involving conceptualization, design, interpretation, drafting of the manuscript, and critical revision of substantial intellectual content. All authors have reviewed and approved the manuscript in its final form.

Conflict of interest

The authors declare no conflict of interest

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References

[1] (a) L. Say, D. Chou, A. Gemmill, Ö. Tunçalp, A. B. Moller, J. Daniels, A. M. Gülmezoglu, M. Temmerman, L. Alkema. Global causes of maternal death: a WHO systematic analysis, *Lancet Glob. Health*, **2014**, *2*, 323-333. [Crossref], [Google Scholar], [Publisher], (b) A. Sadek, R. Baban, M.F. Al-Habib, E.A. Khazaali, Assessment of serum afamin in patients with preeclampsia at third trimester, *Journal of Medicinal and Pharmaceutical Chemistry Research*, **2021**, *3*, 622-626. [Pdf], [Publisher]

[2] L.C. Poon, L.A. Magee, S. Verlohren, A. Shennan, P. von Dadelszen, E. Sheiner, E. Hadar, G. Visser, F. Da Silva Costa, A. Kapur, F. McAuliffe, A literature review and best practice advice for second and third trimester risk stratification, monitoring, and management of pre-eclampsia: Compiled by the Pregnancy and Non-Communicable Diseases Committee of FIGO (the International Federation of Gynecology and Obstetrics), *Int J Gynaecol Obstet* , **2021**, *154*, 3-31. [Crossref], [Google Scholar], [Publisher]

[3] L.A. Magee, M.A. Brown, D.R. Hall, S. Gupte, A. Hennessy, S.A. Karumanchi, L.C. Kenny, F. McCarthy, J. Myers, L.C. Poon, S. Rana, S. Saito, A.C. Staff, E. Tsigas, P. von Dadelszen, The 2021 international society for the study of hypertension in pregnancy classification, diagnosis & management recommendations for international practice, *Pregnancy*

Hypertens, **2022**, *27*, 148-169. [Crossref], [Google Scholar], [Publisher]

[4] M.A. Brown, L.A. Magee, L.C. Kenny, S.A. Karumanchi, F.P. McCarthy, S. Saito, D.R. Hall, C.E. Warren, G. Adoyi, S. Ishaku, Hypertensive disorders of pregnancy: ISSHP classification, diagnosis, and management recommendations for international practice, *Hypertension*, **2018**, *72*, 24-43. [Crossref], [Google Scholar], [Publisher]

[5] S. Rana, E. Lemoine, J. Granger, S.A. Karumanchi, Preeclampsia: pathophysiology, challenges, and perspectives. *Circulation research*, **2019**, *124*, 1094-1112. [Crossref], [Google Scholar], [Publisher]

[6] S. Agrawal, S. Shinar, A.S. Cerdeira, C. Redman, M. Vatish, Predictive performance of PlGF (Placental Growth Factor) for screening preeclampsia in asymptomatic women: a systematic review and meta-analysis, *Hypertension*, **2019**, *74*, 1124-1135. [Crossref], [GoogleScholar], [Publisher]

[7] E.A. Phipps, R. Thadhani, T. Benzing, S.A. Karumanchi, Preeclampsia: pathogenesis, novel diagnostics and therapies, *Nat. Rev. Nephrol*, **2019**, *15*, 275-289. [Crossref], [Google Scholar], [Publisher]

[8] A.C. Staff, The two-stage placental model of preeclampsia: An update, *J. Reprod. Immunol*, **2019**, *134*, 1-10. [Crossref], [Google Scholar], [Publisher]

[9] E. Jung, R. Romero, L. Yeo, N. Gomez-Lopez, P. Chaemsaithong, A. Jaovisidha, F. Gotsch O. Erez, The etiology of preeclampsia, *Am J Obstet Gynecol*, **2022**, *226*, 844-866. [Crossref], [Google Scholar], [Publisher]

[10] X. Bian, A. Biswas, X. Huang, K.J. Lee, T.K. Li, H. Masuyama, A. Ohkuchi, J.S. Park, S. Saito, K.H. Tan, T. Yamamoto, Short-term prediction of adverse outcomes using the sFlt-1 (soluble fms-Like tyrosine kinase 1)/PlGF (placental growth factor) ratio in Asian women with suspected preeclampsia, *Hypertension*, **2019**, *74*, 164-172. [Crossref], [Google Scholar], [Publisher]

[11] S. Rowson, M. Reddy, D.L. De Guingand, A. Langston-Cox, S.A. Marshall, F. da Silva Costa, K.R. Palmer, Comparison of circulating total sFLT-1 to placental-specific sFLT-1 e15a in womenwith suspected preeclampsia. *Placenta*, **2022**, *120*, 73-78. [Crossref], [Google Scholar], [Publisher]

[12] S. Verlohren, S.P. Brennecke, A. Galindo, S.A. Karumanchi, L.B. Mirkovic, D. Schlembach, H. Stepan, M. Vatish, H. Zeisler, S. Rana, Clinical interpretation and implementation of the sFlt-1/PlGF ratio in the prediction, diagnosis and management of preeclampsia, *Pregnancy Hypertension*, **2022**, *27*, 42-50. [Crossref], [Google Scholar], [Publisher]

[13] H. Stepan, A. Galindo, M. Hund, D. Schlembach, J. Sillman, D. Surbek, M. Vatish, Clinical utility of sFlt-1 and PlGF in screening,prediction, diagnosis and monitoring of preeclampsia and fetal growth restriction, *Ultrasound in Obstetrics & Gynecology*, **2023**, *61*, 168-180. [Crossref], [Google Scholar], [Publisher]

[14] L.A. Magee, K.H. Nicolaides, P. von Dadelszen, Preeclampsia, *N Engl J Med*, **2022**, *386*, 1817-1832. [Crossref], [Google Scholar], [Publisher]

[15] S. Rana, S.D. Burke, S.A. Karumanchi, Imbalances in circulating angiogenic factors in the pathophysiology of preeclampsia and related disorders, *Am J Obstet Gynecol*, **2022**, *226*, 1019-1034. [Crossref], [Google Scholar], [Publisher]

[16] T. Yamazaki, A.S. Cerdeira, S. Agrawal, I. Koh, J. Sugimoto, M. Vatish, Y. Kudo, Predictive accuracy of soluble FMS-like tyrosine kinase-1/placental growth factor ratio for preeclampsia in Japan: A systematic review, *Hypertension Research in Pregnancy*, **2021**, *9*, 1-7. [Crossref], [Google Scholar], [Publisher]

[17] A. Umapathy, L.W. Chamley, J.L. James, Reconciling the distinct roles of angiogenic/anti-angiogenic factors in the placenta and maternal circulation of normal and pathological pregnancies, *Angiogenesis*, **2020**, *23*, 105-117. [Crossref], [Google Scholar], [Publisher]

[18] R. Verteramo, M. Pierdomenico, P. Greco C. Milano, The role of melatonin in pregnancy

and the health benefits for the Newborn, *Biomedicines*, **2022**, *10*, 3252. [Crossref], [Google Scholar], [Publisher]

[19] I. Fantasia, S. Bussolaro, T. Stampalija, D. L. Rolnik. The role of melatonin in pregnancies complicated by placental insufficiency: A systematic review, *European Journal of Obstetrics & Gynecology and Reproductive Biology*, **2022**, *278*, 22-28. [Crossref], [Google Scholar], [Publisher]

[20] A. Langston-Cox, S. A. Marshall, D. Lu, K. R. Palmer, E.M. Wallace, Melatonin for the management of preeclampsia: A review, *Antioxidants*, **2021**, *10*, 376. [Crossref], [Google Scholar], [Publisher]

[21] N.A.S. Mohd Azmi, N. Juliana, S. Azmani, N. Mohd Effendy, I.F. Abu, N.I. Mohd Fahmi Teng S. Das, Cortisol on circadian rhythm and its effect on cardiovascular system, *International Journal of Environmental Research and Public Health*, **2021**, *18*, 676 [Crossref], [Google Scholar], [Publisher]

[22] A. Ditisheim, C. Dibner, J. Philippe, A. Pechère-Bertschi, Biological rhythms and preeclampsia, *Frontiers in Endocrinology*, **2013**, *4*, 1-8, Review. [Crossref], [Google Scholar], [Publisher]

[23] S. Bouchlariotou, V. Liakopoulos, M. Giannopoulou, S. Arampatzis, T. Eleftheriadis, P.R. Mertens, E. Zintzaras, I.E. Messinis, I. Stefanidis, Melatonin secretion is impaired in women with preeclampsia and an abnormal circadian blood pressure rhythm, *Renal Failure*, **2014**, *36*, 1001-1007. [Crossref], [Google Scholar], [Publisher]

[24] M.E. Aml, The interaction between maternal and fetal hypothalamic – pituitary – adrenal axes, *Corticosteroids*, P. Celso Ed.; IntechOpen, **2021**, 1-12. [Crossref]

[25] L. Saleh, A.H. van den Meiracker, R. Geensen, A. Kaya, J.E. Roeters van Lennep, J.J. Duvekot, K. Verdonk, E.A.P. Steegers, H. Russcher, A.H.J. Danser, Soluble fms-like tyrosine kinase-1 and placental growth factor kinetics during and after pregnancy in women with suspected or confirmed preeclampsia, *Ultrasound in Obstetrics & Gynecology*, **2018**, *51*, 751-757. [Crossref], [Google Scholar], [Publisher]

[26] S. Rana, S. Salahuddin, A. Mueller, A.H. Berg, R.I. Thadhani, S.A. Karumanchi, Angiogenic biomarkers in triage and risk for preeclampsia with severe features, *Pregnancy Hypertension*, **2018**, *13*, 100-106. [Crossref], [Google Scholar], [Publisher]

[27] M.H. Bublitz, G. Bourjeily, C. D'Angelo, L. R. Stroud, Maternal sleep quality and diurnal cortisol regulation over pregnancy, *Behavioral Sleep Medicine*, **2018**, *13*, 100-106. [Crossref], [Google Scholar], [Publisher]

[28] Roche Diagnostics, *Elecsys sFlt-1*; Roche Diagnostics GmbH, **2018** [Publisher]

[29] Roche Diagnostics, *Elecsys PlGF* Roche Diagnostics GmbH, **2022**. [Publisher]

[30] J. Huart, A. Persu, J.P. Lengelé, J.M. Krzesinski, F. Jouret, G.S. Stergiou, Pathophysiology of the nondipping blood pressure pattern, *Hypertension*, **2023**, *80*, 719-729. [Crossref], [Google Scholar], [Publisher]

[31] V. Pergialiotis, N. Karampetsou, I. Bellos, N. Papantoniou, G. Daskalakis. Nocturnal blood pressure alterations in patients with preeclampsia- Do they really matter? A systematic review of the literature. *European Journal of Obstetrics and Gynecology and Reproductive Biology*, **2019**, *239*, 39-44. [Crossref], [Google Scholar], [Publisher]

[32] A. B. Diallo, B. Coiffard, R. Desbriere, M. Katsogiannou, X. Donato, F. Bretelle, S. Mezouar, J.L. Mege, Disruption of the expression of the placental clock and melatonin genes in preeclampsia, *International Journal of Molecular Sciences*, **2023**, *24*, 2363 [Crossref], [Google Scholar], [Publisher]

[33] K. Bates, E.D. Herzog, Maternal-fetal circadian communication during pregnancy, *Frontiers in Endocrinology*, **2020**, *11*, 1-9, Review. [Crossref], [Google Scholar], [Publisher]

[34] K. Kosicka, A. Siemiątkowska, A. Szpera-Goździewicz, M. Krzyścin, G.H. Bręborowicz, F.K. Główka, Increased cortisol metabolism in women with pregnancy-related hypertension,

Endocrine, **2018**, *61*, 125-133. [Crossref], [Google Scholar], [Publisher]

[35] T.G.R. Húngaro, M.F. Gregnani, T. Alves-Silva, F. Herse, N. Alenina, M. Bader, R.C. Araújo, Cortisol dose-dependently impairs migration and tube-like formation in a trophoblast cell line and modulates inflammatory and angiogenic genes, *Biomedicines*, **2021**, *9*, 980. [Crossref], [Google Scholar], [Publisher]

[36] S. Chan, M. Debono, Review: replication of cortisol circadian rhythm: new advances in hydrocortisone replacement therapy, *Therapeutic Advances in Endocrinology and Metabolism*, **2010**, *1*, 129-138. [Crossref], [Google Scholar], [Publisher]

[37] G. Wang, Y. Huang, T. Hu, B. Zhang, Z. Tang, R. Yao, Y. Huang, X. Fan, X. Ni. Contribution of placental 11β-HSD2 to the pathogenesis of preeclampsia, *The FASEB Journal*, **2020**, *34*, 15379-15399. [Crossref], [Google Scholar], [Publisher]

[38] K. Kosicka-Noworzyń, A. Siemiątkowska, F. Główka, 11βHydroxysteroid dehydrogenase 2 in preeclampsia, *International Journal of Endocrinology*, **2016**, *2016*, 1-9. [Crossref], [Google Scholar], [Publisher]

[39] F.J. Valenzuela-Melgarejo, C. Lagunas, F. Carmona-Pastén, K. Jara-Medina, G. Delgado, Supraphysiological role of melatonin over vascular dysfunction of pregnancy, a new therapeutic agent? *Frontiers in Physiology*, **2021**, *12* , 1-13. [Crossref], [Google Scholar], [Publisher]

[40] Y. Tang, K. Groom, L. Chamley, Q. Chen, Melatonin, a potential therapeutic agent for preeclampsia, reduces the extrusion of toxic extracellular vesicles from preeclamptic placentae, *Cells*, **2021**, *10*, 1904. [Crossref], [Google Scholar], [Publisher]

[41] R. McCarthy, E.S. Jungheim, J.C. Fay, K. Bates, E.D. Herzog, S.K. England, Riding the rhythm of melatonin through pregnancy to deliver on time, *Frontiers in Endocrinology*, **2019**, *10*, 1-11, Review. [Crossref], [Google Scholar], [Publisher]

[42] M. Miyata, M. Ohishi, Novel strategies for the management of nocturnal hypertension, *Circulation Journal*, **2017**, *81*, 938-939. [Crossref], [Google Scholar], [Publisher]

[43] K.E. Duhig, J.E. Myers, C. Gale, J.C. Girling, K. Harding, A. Sharp, N.A.B. simpson, D. Tuffnell, P.T. Seed, A.H. Shennan, Placental growth factor measurements in the assessment of women with suspected Preeclampsia: A stratified analysis of the PARROT trial, *Pregnancy Hypertension*, **2021**, *23*, 41-47. [Crossref], [Google Scholar], [Publisher]

[44] S. Hoshide, Nocturnal hypertensionsolving the puzzle of preeclampsia risk, *Hypertension Research*, **2021**, *44*, 1681-1682. [Crossref], [Google Scholar], [Publisher]

[45] J. Baker, K. Kimpinski. Role of melatonin in blood pressure regulation: An adjunct anti-hypertensive agent, *Clinical and Experimental Pharmacology and Physiology*, **2018**, *45*, 755-766. [Crossref], [Google Scholar], [Publisher]

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