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Association of maternal mitochondrial DNA copy numbers with preeclampsia

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Abstract

Background: Preeclampsia (PE) is a hypertension disorder of pregnancy that affects 2–10% of pregnancies worldwide, with a particularly high prevalence in Bangladesh (14.4%). While the etiology is unclear, mitochondrial dysfunction and oxidative stress have been suggested by evidence. This study aimed to investigate the association of maternal mitochondrial DNA copy number (mtDNA-CN) and PE in Bangladeshi women.

Methods: Fifty-one preeclamptic patients and 51 age- and gestational age-matched (20–40 weeks) normotensive pregnant controls at Bangabandhu Sheikh Mujib Medical University were taken in this case–control study. Peripheral blood samples were collected for DNA extraction, and mtDNA-CN was quantified by quantitative polymerase chain reaction based on the ratio of the mitochondrial NADH dehydrogenase-1 gene and nuclear hemoglobin subunit β gene. Optimal cut-off points were determined by receiver operating characteristic (ROC) analysis, and odds ratios (ORs) were calculated to assess PE risk.

Results: Preeclamptic women had significantly higher mean mtDNA-CN (82.1 ± 80.3) than controls (44.7 ± 25.5, $P = 0.002$) with greater heterogeneity among the PE group (range: 11.0–404.5 vs. 4.3–128.8). In ROC analysis, the area under the curve was 0.688 ($P < 0.001$) with an optimal cut-off >45, which was equivalent to 68.6% sensitivity and 58.8% specificity. Women with mtDNA-CN >45 had three-fold greater PE odds (OR = 3.1, 95% CI: 1.4–7.0, $P = 0.005$), with an overall diagnostic accuracy of 63.7%.

Conclusion: This investigation exhibited a strong link between high maternal mtDNA-CN and PE in Bangladeshi women. The results implicate mitochondrial dysfunction in PE pathophysiology and suggest the usefulness of mtDNA-CN as a biomarker. Studies involving larger populations are necessary to confirm the findings and determine mtDNA-CN's predictive potential before the clinical onset of PE.

Keywords: Bangladesh, biomarker, mitochondrial DNA copy number, oxidative stress, preeclampsia

Introduction

Preeclampsia (PE) is a pregnancy-related multisystem disease characterized by the recent onset of hypertension (systolic blood pressure [BP] ≥140 mmHg and diastolic BP ≥90 mmHg) with proteinuria or end-organ damage after 20 weeks of pregnancy, with secondary renal insufficiency, hepatic dysfunction, hematologic findings, neurological disorder, or uteroplacental dysfunction.[1] PE is a severe global health issue, affecting 2–10% of pregnancies worldwide and

responsible for a significant contribution to maternal and perinatal morbidity and mortality.[2] PE and eclampsia are responsible for more than 50,000 deaths among women annually, with an overrepresentation among African-American and Hispanic women.[3] The burden is particularly significant in developing countries, with PE accounting for an estimated 60,000 maternal deaths annually.[4] The difference is dramatic between the high-income and low-income countries, with PE being responsible for 0.4% of maternal deaths in high-income countries but 2.8% in low-income countries.[5] It was 19/1,000 births in Thailand in 2017,[6] and in Bangladesh, it is comparatively high at 14.4%.[7] The incidence is also parity-dependent at 4.1% in nulliparous and 1.7% in multiparous women.[8] In addition to pregnancy-related morbidities, PE has serious long-term health implications in terms of increased lifetime risk for cardiovascular disease.[9] The maternal complications include eclampsia, HELLP syndrome, renal failure, coagulopathy, placental abruption, and cerebrovascular accident.[10] Fetal complications include growth restriction, preterm labor, and perinatal mortality augmentation with PE-caused growth restriction occurring in 22.2% of such pregnancies.[11] The pathophysiology of PE is based on placental dysfunction. In normal pregnancy, cytotrophoblast invasion of maternal spiral arteries to form vascular sinuses is coupled with the progression of invasion into the myometrium and remodeling of maternal vessels into high-capacity, high-flow conduits.[12] Though the precise processes are unknown, PE pathogenesis is most likely associated with insufficient trophoblastic invasion and differentiation.[13] Oxidative stress is increasingly being recognized as one of the leading etiological determinants of PE development.[14] Placental ischemia-reperfusion injury during early pregnancy increases oxidative stress, which can be recognized as early as 8–10 weeks gestation.[15] Oxidative stress affects placental and maternal circulation, resulting in vascular dysfunction.[14] Mitochondrial dysfunction, particularly within trophoblastic cells, appears to be critical in the process, which prevents normal development of the placenta.[16] Mitochondrial oxidative phosphorylation provides the main energy source for placental function and fetal growth.[17] Dysfunction of the placental mitochondrial respiratory chain complex increases the production of reactive oxygen species and oxidative stress, possibly implicated in PE pathogenesis.[15] Various biomarkers have been investigated for the prediction of PE, including angiogenic markers (vascular endothelial growth factor, placental growth factor, soluble Fms-like tyrosine kinase-1), immunologic (Placental Protein 13, pregnancy-associated plasma protein A), and endocrine (activin-A and inhibin-A) markers.[18,19] Among these, mtDNA-CN was identified as a new marker of oxidative stress and mitochondrial dysfunction.[18] Maternal quantifies mitochondrial DNA copy number (mtDNA-CN) in cells and differs from nuclear DNA under exclusive maternal transmission.[20] It participates in cellular energy generation and is increasingly investigated for its role in various states of disease.[21] Elevated levels of mtDNA-CN typically indicate disrupted mitochondrial function in conjunction with disease states.[22] The measurement of mtDNA-CN is very instructive to learn about PE etiology since mtDNA is extremely susceptible to oxidative damage[23] and mitochondrial metabolism also changes dramatically during pregnancy.[22] Nonetheless, there is a vast information emptiness that has to be addressed by this study because relatively few studies have shown the alteration in mtDNA-CN in PE[18] and none have examined the connection between the two in Bangladeshi women.

Methods

This is a case–control study that explored the association between maternal mtDNA-CN and PE at Bangabandhu Sheikh Mujib Medical University in Bangladesh over 1 year. Using purposive sampling, the study recruited 51 PE cases from the Feto-maternal Medicine department and 51 normotensive healthy pregnant women as controls from the Obstetrics and Gynecology department. Sample size was calculated by two-sample t-tests formula with unequal variance, with 80% power to detect a population mean difference of 1.0

between groups (mean mtDNA-CN: case = 3.0, control = 2.0) with standard deviations of 1.5 for both groups at a 0.05 significance level. Inclusion criteria for cases required systolic BP ≥140 mmHg and/or diastolic BP ≥90 mmHg, proteinuria or organ involvement, and gestational age of 20–40 weeks. Controls were applied as having systolic BP <140 mmHg, diastolic BP <90 mmHg, no proteinuria, and gestational age 20–40 weeks. The following patients were excluded: those with multiple pregnancies, chronic hypertension, renal disease, diabetes mellitus, antiphospholipid antibody syndrome, malignancy, autoimmune or neurodegenerative diseases, intrauterine death, or known congenital anomalies. The data gathered were socio-demographic information, obstetric history, physical examination, and 3ml blood samples from the antecubital vein. Genomic DNA was extracted from maternal peripheral blood leukocytes, and mtDNA-CN was quantitated by modified quantitative polymerase chain reaction (qPCR) through the quantitation of the ratio of mitochondrial-encoded NADH dehydrogenase-1 to the nuclear gene hemoglobin subunit β. Duplicate samples were analyzed with three quality controls per plate by blinded scholars to clinical data and disease status. Statistical examination was done on SPSS version 26 using descriptive statistics, Chi-square tests for categorical data, unpaired t-tests for continuous data, receiver operating characteristic (ROC) curve analysis to determine the optimal cut-off value of mtDNA-CN, and odds ratio (OR) determination with 95% confidence intervals to assess PE risk. The study was ethically cleared by the Institutional Review Board with informed consent from all participants.

Results

Table 1 emphasizes a comprehensive breakdown of the socio-demographic profile of the study sample. The age distribution displays that most participants in both groups (49.0%) were 21–30 years old, which corresponds to women of childbearing age. While the mean age was slightly higher in cases (30.0 ± 4.3 years) than in controls (28.5 ± 4.8 years), this difference was not statistically significant

Table 1: Socio-demographic the participants (*n*=102)

Characteristics	Case *n* (%)	Control *n* (%)	*P*-value
Age (years)			
≤20	3 (5.9)	5 (9.8)	0.744[a]
21–30	25 (49)	25 (49.0)	
>30	23 (45.1)	31 (41.2)	
Mean±SD	30.0±4.3	28.5±4.8	
Educational qualification			
Illiterate	4 (7.8)	3 (5.9)	0.063[a]
Primary	6 (11.8)	8 (15.7)	
SSC	6 (11.8)	17 (33.3)	
HSC	35 (68.6)	23 (45.1)	
Monthly family income (BDT)			
<10,000	2 (3.9)	3 (5.9)	0.281[a]
10,000-25,000	17 (33.3)	24 (47.1)	
>25,000	32 (67.7)	24 (47.1)	
Total	51 (100)	51 (100)	

[a]*P*-value obtained from Chi-square test, HSC: Higher secondary education, SSC: Secondary school education

($P > 0.05$), reflecting successful matching of ages between groups. The educational background was also very dissimilar, with cases having a much higher proportion of those with higher secondary education (68.6% vs. 45.1% in controls), although this was on the verge of but not statistically significant ($P = 0.063$). This recommends that women with PE in this population may have had better access to education or even better health literacy, which could influence healthcare-seeking behavior. Economic status, as monthly family income, identified two-thirds of the cases (67.7%) with over 25,000 BDT per month to the controls' less than half (47.1%). While not statistically significant ($P = 0.281$), the trend raises speculation as to whether socioeconomic factors may have a part to play in determining rates of detection of PE, perhaps through increased access to antenatal care or by differences in lifestyle.

The numerous obstetric profiles that together make up the basic baseline characteristics required to carry out an exhaustive and exacting study on pregnancy and its related aspects are

comprehensively and thoroughly outlined in Table 2. The first pregnancy (primigravida) versus multiple pregnancies (multigravida) status distribution showed comparable proportions in cases (27.5% primigravida) and controls (31.4% primigravida) with no statistically significant difference ($P = 0.664$). This is important because uniformity is an established PE risk factor, and women with their first pregnancy are typically more at risk. The comparable distribution means that differences in outcome that are observed are less likely to be confounded by parity status. Distribution of gestational age is significant background information for the results since PE presentation and pathophysiology can shift with advancing pregnancy. The majority of participants in both groups were in the third trimester (31–36 weeks) with 72.5% of the cases and 62.7% of the controls falling within that group. No subjects were engaged into the earliest gestational age category (20–25 weeks), compared to one control subject (2.0%), as anticipated by the typical clinical presentation pattern of PE, which becomes more common in late pregnancy. The similar breakdown within gestational age groups ($P = 0.333$) implies adequate matching between groups, adding to the validity of comparisons of mtDNA-CN levels. This pairing is of special importance because the gestational age can influence mitochondrial parameters irrespective of PE status.

Table 2: Obstetrics characteristics of the participants (n=102)

Characteristics	Case *n* (%)	Control *n* (%)	*P*-value
Gravida			
Primigravida	14 (27.5)	16 (31.4)	0.664[a]
Multigravida	37 (72.5)	35 (68.6)	
Gestational age (weeks)			
20–25	0 (0)	1 (2)	0.333[a]
26–30	4 (7.8)	9 (17.6)	
31–36	37 (72.5)	32 (62.7)	
>36	10 (19.6)	9 (17.6)	
Total	51 (100)	51 (100)	

[a]*P*-value obtained from Chi-square test

A representation of the Mean mtDNA-CN in the participants is shown in Figure 1. Patients with PE have a markedly higher mtDNA-CN. The bar graph provides a visual representation of the stark contrast in mean mtDNA-CN in controls (44.7) versus cases (82.1), with error bars likely to be a standard error of the mean. This nearly twofold increase in mtDNA-CN in preeclamptic women shows extremely important alterations in mitochondrial dynamics and potentially enormous mitochondrial stress or mitochondrial dysfunction in the pathophysiology of PE.

The data from Table 3 provides a comparison of mtDNA-CN between case and control. The standard deviation in the PE group (80.3) was distinctly higher compared with controls (25.5), indicating higher heterogeneity in mitochondrial DNA content among women with PE. Such heterogeneity can be the result of different pathophysiological mechanisms, disease severities, or response patterns in preeclamptic women. The broad distribution within the PE group (11.0–404.5) versus controls (4.3–128.8) reinforces this heterogeneity and implies that some preeclamptic women had profoundly raised levels of mtDNA-CN. That the difference between groups is statistically significant ($P = 0.002$) means that such results are highly unlikely to have occurred by chance and lends support to the hypothesis that mitochondrial disease, as

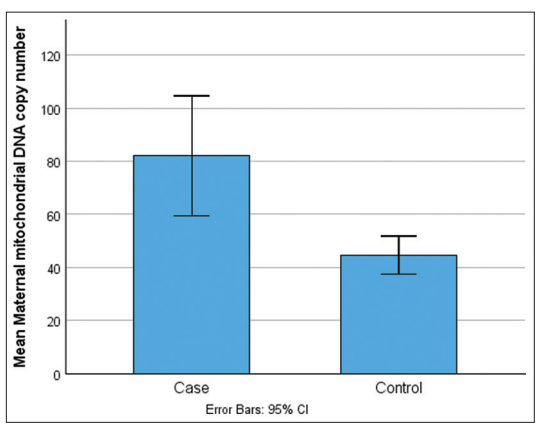

Figure 1: Mean mitochondrial DNA copy number in the participants

indicated by disturbed mtDNA-CN, is linked with PE.

Figure 2 illustrates a ROC curve comparison of the discriminatory power of mtDNA-CN for PE diagnosis. The smooth curve graphically presents sensitivity (true positive rate) versus 1-specificity (false positive rate) at various possible diagnostic cut-points. The area under the curve (AUC) value of 0.688 signifies moderate discriminatory power, and values ranging from 0.7 to 0.8 are generally considered sufficient for biomarker performance. The statistically higher AUC ($P < 0.001$) also suggests that mtDNA-CN performs better than at random (the diagonal reference line from the start to the top right would mark this, so an AUC of 0.5). The curve tells us about sensitivity against specificity trade-offs at different cut-off levels; the optimal threshold is typically at the one where the vertical distance from the diagonal reference line is maximized. While not passing the cutoff of being an excellent diagnostic test (usually, AUC >0.8), this is promising in a single marker and

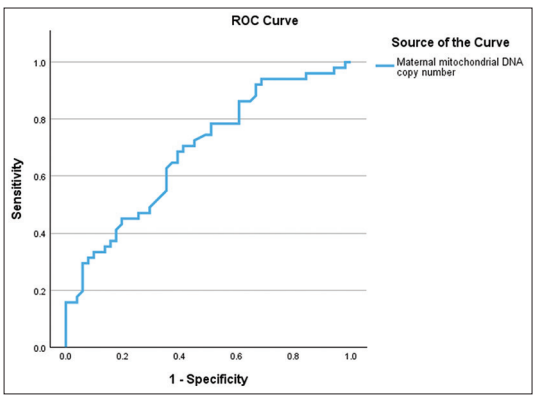

Figure 2: Receiver operating characteristic analysis of maternal mitochondrial DNA copy number in case and control

Table 3: Comparison of mitochondrial DNA copy number between case and control (n=102)

Group	*n*	Mean	SD	Min-Max	*P*-value
Case	51	82.1	80.3	11.0–404.5	0.002[b]
Control	51	44.7	25.5	4.3–128.8	

[b]*P*-value obtained from unpaired t-test

suggests that mtDNA-CN might provide valuable information to PE risk assessment, particularly if combined with other clinical and biochemical markers in a multi-marker panel.

Table 4 provides the information compulsory to pick the best cut-off for clinical application by comparing the diagnostic performance characteristics of mtDNA-CN at multiple possible threshold values. For each potential cut-off, the table provides the resultant sensitivity (ratio of true PE cases correctly classified), specificity (ratio of true non-PE cases correctly classified), and Youden Index (sensitivity + specificity −1), which is an index of the overall discriminatory ability at that cut-off. There is an increasing trade-off between sensitivity and specificity as the cut-off increases. For lower thresholds (e.g., >20), sensitivity is extremely high (0.96), but so is the false-positive rate (0.14), so virtually all instances of PE would be detected, but with an overwhelming number of false positives. Increasing the threshold reduces sensitivity but increases specificity. With the Youden Index, which considers these two opposing factors, an optimal cut-off value of >45 was determined with a sensitivity of 0.69 and specificity of 0.61, with the highest Youden Index

Table 4: Sensitivity and specificity of maternal mitochondrial DNA copy number in different cut-off values

Cut-off value	Sensitivity	Specificity	Youden Index (Sensitivity+ Specificity−1)
>20	0.96	0.14	0.10
>25	0.94	0.24	0.18
>30	0.92	0.31	0.24
>35	0.84	0.39	0.24
>40	0.77	0.49	0.26
>45	0.69	0.61	0.29
>50	0.59	0.65	0.24
>55	0.45	0.80	0.26
>60	0.37	0.82	0.20
>65	0.33	0.86	0.20
>70	0.33	0.90	0.24

Table 5: Association between maternal mitochondrial DNA copy number and preeclampsia (n=102)

Mitochondrial DNA copy number	Case n (%)	Control n (%)	P-value	OR (95% CI)
>45	35 (68.6)	21 (41.2)	0.005[a]	3.1 (1.4–7.0)
≤45	16 (31.4)	30 (58.8)		
Total	51 (100)	51 (100)		

[a]P-value obtained from Chi-square test

of 0.29. This clarifies the considerations apposite to determining the diagnostic threshold and enables healthcare practitioners to comprehend the ramifications associated with using mtDNA-CN as a diagnostic assay.

Table 5 assigns a quantitative value to the strength of the relationship between raised mtDNA-CN and PE, with the generally accepted optimum cut-off value of >45. The values illustrate that more than two-thirds of women with PE (68.6%) had values above this cut-point, compared to only 41.2% of controls. The proportion difference was statistically significant (P = 0.005), providing conclusive proof of association. The OR of 3.1, with a 95% confidence interval of 1.4–7.0, offers a quantitation of this association, and from it, one can observe that women with mtDNA-CN >45 had approximately three times higher odds of getting PE than with less than these values.

Table 6 represents an overall appraisal of the diagnostic precision of mtDNA-CN with a cut-off of >45 as established. This 68.6% sensitivity (95% CI 54.1–80.9%) implies that this cut-off would recognize roughly two for every three PE women but miss roughly a third (false negatives). With the specificity being 58.8% (95% CI 44.2–72.4%), this would imply that roughly 60% of non-PE women would correctly be diagnosed negative and roughly 40% of them would mistakenly be reported positive. The positive predictive value of 62.5% (95% CI 53.3–70.8%) is that 63% of women with mtDNA-CN >45 would have PE. The negative predictive value of 65.2% (95% CI 54.1–74.9%) is that among women with mtDNA-CN ≤45, 65% would not have PE. The overall accuracy of 63.7% (95% CI 53.6–73.0%) is the proportion of all women (both with and without PE) who would be correctly classified by using this threshold.

Table 6: Diagnostic accuracy of serum mtDNA-CN level to identify preeclampsia

Statistics	Value (%)	95% CI
Sensitivity	35/51=68.6	54.1–80.9
Specificity	30/51=58.8	44.2–72.4
PPV	35/56=62.5	53.3–70.8
NPV	30/46=65.2	54.1–74.9
Accuracy	(35+30)/102=63.7	53.6–73.0

PPV: Positive predictive value, NPV: Negative predictive value, mtDNA-CN: Mitochondrial DNA copy number

Discussion

This study revealed a significant association between elevated maternal mtDNA-CN and PE in a Bangladeshi population. PE patients had nearly two-fold higher mean mtDNA-CN (82.1) compared to normotensive controls (44.7) (P = 0.002), suggesting extreme mitochondrial dysregulation in the etiology of PE. The significantly greater mtDNA-CN in the PE group is in line with some previous studies favoring mitochondrial dysfunction in PE pathogenesis. Williamson *et al*.[24] reported similar results, with increased mtDNA-CN in the placental tissue of PE patients, which they attributed to compensatory mitochondrial biogenesis due to oxidative stress. Increased mtDNA-CN would therefore be a sign of cellular adaptation to mitochondrial dysfunction and increased oxidative stress, where mitochondria attempt to compensate for dysfunctional capacity by increasing their number.[25] The wide variability of mtDNA-CN among PE cases (11.0–404.5) compared to controls (4.3–128.8) reflects heterogeneity in mitochondrial response patterns.

This variability could be due to differences in pathophysiologic processes or disease severities in the PE spectrum, as Vishnyakova *et al*.[26] hypothesized, where differences in mtDNA-CN correlated with clinical severity. ROC analysis by us yielded an AUC of 0.688, reflecting a moderate diagnostic value of mtDNA-CN as a biomarker for PE. The optimum cut-off of >45 provided matched sensitivity (68.6%) and specificity (58.8%), with a three-fold increase in PE risk (OR = 3.1, 95% CI 1.4–7.0) above this threshold. These figures demonstrate mtDNA-CN's clinical utility but also highlight its restrictions as a single diagnostic marker. Holland *et al*.[17] also concluded that mitochondrial markers must be integrated into multi-parameter models to improve PE prediction. This study contributes to PE pathophysiology knowledge from a mitochondrial point of view. The association between elevated mtDNA-CN and PE is proof in favor of the growing body of literature pointing to placental dysfunction and disturbed mitochondrial function.[15] Mitochondria, being the primary cellular energy producers and modulators of oxidative stress, have significant roles in placental development and function. Their malfunction possesses the potential to cause oxidative damage, inflammation, and endothelial damage, hallmark features in PE pathogenesis.[14] Compared to the previous studies that were focused on placental tissue, our study with maternal peripheral blood demonstrates that systemic mitochondrial dysregulation is identifiable in accessible maternal samples. This finding is consistent with Qiu *et al*.,[27] who reported that maternal circulatory biomarkers reflect placental disturbances and can serve as non-invasive markers of pregnancy complications. Despite some limitations, our findings show a significant correlation between maternal mtDNA-CN and PE among the Bangladeshi population. The moderate diagnostic precision of mtDNA-CN suggests clinical value, particularly within resource-constrained settings, such as Bangladesh, where PE causes a considerable fraction of maternal fatalities.[3] Furthermore, clarifying the mitochondrial role in the pathogenesis of PE might contribute to the planning of novel focused therapeutic measures designed to augment mitochondrial function and limit oxidative damage.[16] A subsequent study will explore longitudinal changes in mtDNA-CN across pregnancy to validate its predictive value before the clinical onset of PE. In addition, the evaluation of the correlation between mtDNA-CN and PE severity, along with incorporation with other established known biomarkers, will further enhance its clinical use within combined risk prediction models.

Limitations of the study

This study recruited a relatively small sample size, which is also a limitation of the present study. Therefore, in the future, further study may be undertaken with a large sample size.

Conclusion

This study establishes a useful association of elevated maternal mtDNA-CN with PE in Bangladeshi women, involving mitochondrial dysfunction in PE pathophysiology. While demonstrating modest diagnostic accuracy (AUC = 0.688) with a three-fold increased risk for PE compared to the optimum cut-off value of >45, mtDNA-CN shows potential as a biomarker for PE risk. These findings contribute to the building body of knowledge on the pathogenesis of PE and indicate the promise of mitochondria-targeted therapies for this life-threatening pregnancy complication that continues to exact a significant impact on maternal and fetal outcomes worldwide.

Recommendations

Large population-based and advanced studies should be conducted, taking into account all confounding variables. The diagnostic effectiveness of mtDNA-CN in detecting PE has the potential to lower maternal morbidity and mortality. This emphasizes the necessity of additional study and the possible use of mtDNA-CN as a diagnostic tool to improve maternal health outcomes. This study establishes a link between maternal mtDNA-CN and PE, which contributes to a better knowledge of the etiopathogenesis of PE

and offers up the possibility of further study to determine this biomarker as a predictor of PE. Further study would establish the critical role of mitochondrial abnormalities in placental and endothelial dysfunction, making targeting mitochondria to improve or restore mitochondrial function a promising therapeutic option.

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Conflict of Interest

None declared.

References

1. Overton E, Tobes D, Lee A. Preeclampsia diagnosis and management. Best Pract Res Clin Anaesthesiol 2022;36:107-21.
2. Poon LC, Shennan A, Hyett JA, Kapur A, Hadar E, Divakar H, *et al*. The International Federation of Gynecology and Obstetrics (FIGO) initiative on preeclampsia (PE): A pragmatic guide for first trimester screening and prevention. Int J Gynaecol Obst 2019;145:1-33.
3. Begum MR, Begum A, Quadir E, Akhter S, Shamsuddin L. Eclampsia: Still a problem in Bangladesh. MedGenMed 2004;6:52.
4. Pasha O, McClure EM, Saleem S, Tikmani SS, Lokangaka A, Tshefu A, *et al*. A prospective cause of death classification system for maternal deaths in low and middle□income countries: Results from the Global Network Maternal Newborn Health Registry. BJOG 2018;125:1137-43.
5. Madise N, Nandi A, Wong B, Razvi S. Achieving maternal and neonatal mortality development goals effectively: A cost-benefit analysis. J Benefit Cost Anal 2023;14:206-34.
6. Chantanavilai S, Kongwattanakul K, Komwilaisak R, Saksiriwuttho P, Chaiyarach S, Duangkam C, *et al*. Epidemiology and prognostic factors for successful expectant management of early-onset severe features preeclampsia: A retrospective multicenter cohort study. Pregnancy Hypertens 2022;30:226-31.
7. Mou AD, Barman Z, Hasan M, Miah R, Hafsa JM, Das Trisha A, *et al*. Prevalence of preeclampsia and the associated risk factors among pregnant women in Bangladesh. Sci Rep 2021;11:21339.
8. Hernández-Díaz S, Toh S, Cnattingius S. Risk of pre-eclampsia in first and subsequent pregnancies: Prospective cohort study. BMJ 2009;338:b2255.
9. Ciumărnean L, Milaciu MV, Negrean V, Orăşan OH, Vesa SC, Sălăgean O, *et al*. Cardiovascular risk factors and physical activity for the prevention of cardiovascular diseases in the elderly. Int J Environ Res Public Health 2021;19:207.
10. Kongwattanakul K, Saksiriwuttho P, Chaiyarach S, Thepsuthammarat K. Incidence, characteristics, maternal complications, and perinatal outcomes associated with preeclampsia with severe features and HELLP syndrome. Int J Womens Health 2018;10:371-7.
11. Suemitsu C, Fudaba M, Kitada K, Kurihara Y, Tahara M, Hamuro A, *et al*. Changes of coagulation and fibrinolytic status detected by thromboelastography (TEG6s®) in pregnancy, labor, early postpartum, postpartum hemorrhage and heparin treatment for perinatal venous thrombosis. Healthcare (Basel) 2022;10:2060.
12. Dimitriadis E, Rolnik DL, Zhou W, Estrada-Gutierrez G, Koga K, Francisco RP, *et al*. Pre-eclampsia. Nat Rev Dis Primers 2023;9:8.
13. Harmon AC, Cornelius DC, Amaral LM, Faulkner JL, Cunningham MW Jr., Wallace K, *et al*. The role of inflammation in the pathology of preeclampsia. Clin Sci 2016;130:409-19.
14. Sánchez-Aranguren LC, Prada CE, Riaño-Medina CE, Lopez M. Endothelial dysfunction and preeclampsia: role of oxidative stress. Front Physiol 2014;5:372.
15. Schoots MH, Gordijn SJ, Scherjon SA, van Goor H, Hillebrands JL. Oxidative stress in placental pathology. Placenta 2018;69:153-61.
16. Muralimanoharan S, Maloyan A, Myatt L. Mitochondrial function and glucose metabolism in the placenta with gestational diabetes mellitus: Role of miR-143. Clin Sci 2016;130:931-41.
17. Holland O, Nitert MD, Gallo LA, Vejzovic M, Fisher JJ, Perkins AV. Placental mitochondrial function and structure in gestational disorders. Placenta 2017;54:2-9.
18. Malik AN. Mitochondrial DNA-novel mechanisms of kidney damage and potential biomarker. Curr Opin Nephrol Hypertens 2023;32:528-36.
19. Staff AC. Circulating predictive biomarkers in preeclampsia. Pregnancy Hypertens 2011;1:28-42.
20. Cerantonio A, Citrigno L, Greco BM, De Benedittis S, Passarino G, Maletta R, *et al*. The role of mitochondrial copy number in neurodegenerative diseases: Present insights and future directions. Int J Mol Sci 2024;25:6062.
21. Zhang R, Du J, Xiao Z, Jiang Y, Jin L, Weng Q. Association between the peripartum maternal and fetal telomere lengths and mitochondrial DNA copy numbers and preeclampsia: A prospective case-control study. BMC Pregnancy Childbirth 2022;22:483.

22. Zhao D, Bartz TM, Sotoodehnia N, Post WS, Heckbert SR, Alonso A, *et al*. Mitochondrial DNA copy number and incident atrial fibrillation. BMC Med 2020;18:1-1.
23. Beckman KB, Ames BN. Endogenous oxidative damage of mtDNA. Mutat Res 1999;424:51-8.
24. Williamson RD, McCarthy FP, Kenny LC, McCarthy CM. Activation of a TLR9 mediated innate immune response in preeclampsia. Sci Rep 2019;9:5920.
25. Malik AN, Czajka A. Is mitochondrial DNA content a potential biomarker of mitochondrial dysfunction? Mitochondrion 2013;13:481-92.
26. Vishnyakova PA, Volodina MA, Tarasova NV, Marey MV, Tsvirkun DV, Vavina OV, *et al*. Mitochondrial role in adaptive response to stress conditions in preeclampsia. Sci Rep 2016;6:32410.
27. Qiu C, Hevner K, Enquobahrie DA, Williams MA. Maternal serum heme-oxygenase-1 (HO-1) concentrations in early pregnancy and subsequent risk of gestational diabetes mellitus. PLoS One 2012;7:e48060.