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Short Communication

Cortisol and pregnancy-related anxiety in relation to preeclampsia among third-trimester pregnant women: A case–control study from Aceh, Indonesia

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Abstract

Despite extensive research, the interplay between hormonal stress markers and pregnancy-specific anxiety in the pathogenesis of preeclampsia remains insufficiently understood. This study aimed to analyze the simultaneous relationships between serum cortisol levels, pregnancy-related anxiety, and the occurrence of preeclampsia among third-trimester pregnant women. A case–control study was conducted at three hospitals in Banda Aceh, Indonesia, involving third-trimester pregnant women. Serum cortisol concentrations were measured using an immunoassay, and anxiety levels were assessed with the validated Pregnancy Related Anxiety Questionnaire (PRAQ). Preeclampsia diagnosis followed standard clinical and laboratory criteria. Associations between cortisol levels (categorized into high and normal) and PRAQ scores (classified as high and low–moderate) with preeclampsia were evaluated using the Chi-squared test, and crude odds ratios (OR) with 95% confidence intervals (95%CI). The Spearman's correlation was used to determine the correlation between cortisol levels and PRAG scores. A total of 66 pregnant women were included in the final analysis (33 with preeclampsia and 33 with normal pregnancy). Women with high serum cortisol levels had a markedly greater likelihood of developing preeclampsia compared with those with normal cortisol levels (odds ratio (OR)=34.00; 95% confidence interval (95%CI): 4.93–234.46). Similarly, women with high pregnancy-related anxiety exhibited a significantly elevated risk of preeclampsia (OR=16.71; 95%CI: 4.95–56.39). No significant correlation was observed between cortisol levels and PRAQ scores in both groups (preeclampsia: r=-0.041, p=0.821; normal pregnancy: r=0.278, p=0.117). In conclusion, elevated serum cortisol and high pregnancy-related anxiety are independently associated with preeclampsia, although not directly correlated with each other. These findings highlight the potential of dual screening for cortisol and pregnancy-specific anxiety as an innovative approach for early identification of women at high risk of preeclampsia.

Keywords: Preeclampsia, screening, anxiety, cortisol, PRAQ

Introduction

Preeclampsia is a multifactorial hypertensive disorder of pregnancy characterized by new-onset hypertension and multi-organ dysfunction occurring after 20 weeks of gestation. It affects approximately 2–8 % of pregnancies globally and remains one of the leading causes of maternal and neonatal morbidity and mortality worldwide [1]. In Indonesia, the prevalence of

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Received: November 10, 2025 - Accepted: December 8, 2025 - Published online: December 11, 2025

preeclampsia is estimated at 5.3 %, representing a major contributor to the maternal mortality rate of 305 per 100.000 live births as reported in the 2023 Indonesian Demographic and Health Survey [2].

Despite extensive research, the precise molecular and physiological mechanisms underlying preeclampsia remain incompletely elucidated. Current evidence implicates placental dysfunction, oxidative stress, and systemic inflammatory activation as key pathogenic pathways [3]. More recent investigations have highlighted the potential contributions of psychological and endocrine factors to the development of preeclampsia; however, substantial knowledge gaps persist in understanding the complex interactions between these biological and psychosocial determinants [4].

Cortisol, the primary glucocorticoid hormone secreted by the adrenal cortex, plays a central role in maintaining metabolic homeostasis and mediating the physiological response to stress. During normal pregnancy, circulating cortisol concentrations progressively increase, reaching their peak during the third trimester with up to a threefold elevation compared to non-pregnant women [5]. This physiological rise facilitates the synthesis of pulmonary surfactant and supports maturation of the fetal cardiovascular and central nervous systems [6]. However, investigations have demonstrated that women with preeclampsia exhibit cortisol levels that exceed the expected physiological range [7].

Psychological factors, particularly pregnancy-related anxiety, have also received increasing attention as potential contributors to the development of preeclampsia. A meta-analysis reported that women experiencing high stress had a 1.5- to 2-fold greater risk of developing preeclampsia compared with those without significant stress exposure [8]. Activation of the hypothalamic–pituitary–adrenal axis in response to pregnancy-related anxiety leads to elevated cortisol secretion, which may aggravate endothelial dysfunction and systemic inflammation, thereby exacerbating the pathophysiological processes underlying preeclampsia [9]. To assess pregnancy-specific anxiety, the Pregnancy Related Anxiety Questionnaire (PRAQ) was developed and validated as a psychometric instrument focusing on concerns related to pregnancy, childbirth, and fetal health. A study demonstrated that the PRAQ possesses strong validity and reliability in identifying pregnant women at elevated risk of pregnancy complications, including preeclampsia [10].

Although separate lines of evidence have established associations between cortisol and preeclampsia, as well as between anxiety and preeclampsia, substantial research gaps remain in understanding the simultaneous interactions among these three variables. Most previous studies have been limited to bivariate analyses, while few have comprehensively examined the triadic relationship between cortisol levels, anxiety, and preeclampsia within an integrated research framework. This study aimed to address these gaps by analyzing the concurrent relationships between cortisol concentration, anxiety level measured using the PRAQ, and the occurrence of preeclampsia among third-trimester pregnant women. This comprehensive analytical approach is expected to provide a more in-depth understanding of the interplay between hormonal and psychological factors in the pathogenesis of preeclampsia.

Methods

Study design and setting

This study employed an observational analytic design with a case–control approach to investigate the relationships between serum cortisol levels and pregnancy-related anxiety with preeclampsia. The study was conducted in 2025 at Dr. Zainoel Abidin Hospital, Rumah Sakit Ibu & Anak Hospital, and Meuraxa General Hospital in Banda Aceh, Aceh Province, Indonesia. These hospitals function as tertiary referral centers for obstetric and perinatal care in the province and manage a significant preeclampsia cases. All eligible participants were screened during routine antenatal visits or during inpatient care, following a consecutive sampling strategy to minimize selection bias and ensure representation of typical patient flow. Before enrollment, each participant received a detailed explanation of the study objectives, procedures, and potential risks. Written informed consent was obtained in accordance with ethical principles outlined in the Declaration of Helsinki. The study setting allowed for standardized clinical evaluation,

controlled timing of blood sampling, and administration of the validated Indonesian version of the PRAQ in a private, confidential environment. Data collection was carried out in the antenatal clinics, obstetrics and gynecology wards, and delivery rooms at each site to ensure recruitment of both preeclampsia cases and normotensive controls with comparable gestational ages.

Sample and characteristics

The study population comprised third-trimester pregnant women (gestational age ≥34 weeks) attending antenatal care clinics, obstetric wards, and delivery units at the participating hospitals. Participants were classified into two groups: (1) a case group consisting of women diagnosed with preeclampsia according to standard clinical and laboratory criteria (blood pressure ≥140/90 mmHg after 20 weeks of gestation with proteinuria or signs of multi-organ involvement); and (2) a control group comprising normotensive pregnant women without obstetric or systemic complications.

Eligible participants were women aged 20–44 years who consented to participate. Exclusion criteria included a history of severe psychiatric illness, use of medications that affect cortisol or anxiety levels (e.g., corticosteroids, antidepressants), presence of chronic hypertension, diabetes mellitus, multiple pregnancy, or fetal abnormalities.

Sample size and sampling method

The sample size was calculated using the unpaired comparative numerical study formula, assuming a 95% confidence level and 80% power. Based on preliminary data and expected correlation differences, a minimum of 30 participants per group was required. To account for potential attrition, 10% was added, yielding a final sample size of 33 participants in each group, totaling 66 subjects. Participants were recruited consecutively from eligible patients presenting to the study sites during the data collection period to ensure representativeness and minimize selection bias.

Study variables and measurements

The independent variables in this study were serum cortisol concentration and pregnancy-related anxiety, while the dependent variable was preeclampsia status. Serum cortisol levels were measured from 2 mL of venous blood collected between 06:00 and 12:00 hours to account for circadian variation. Samples were centrifuged at 2000×g for 10 minutes, and serum aliquots were stored at -80°C until analysis. Cortisol concentrations were quantified using the Elecsys Cortisol immunoassay kit (Roche, Basel, Switzerland) in accordance with the manufacturer's instructions.

Pregnancy-related anxiety was assessed using the validated Indonesian version of the PRAQ, which comprises 10 items rated on a five-point Likert scale. The total score ranges from 10 to 50, with higher scores indicating greater anxiety. Scores were categorized as low (10–19), moderate (20–29), and high (≥30) anxiety levels [10].

Data collection

After obtaining informed consent, participants were interviewed to collect demographic data, obstetric history, and relevant clinical information using a structured questionnaire. Blood pressure measurements and clinical evaluations were conducted by trained obstetric residents under the supervision of an obstetrics and gynecology consultant. Venous blood samples were collected under aseptic conditions, processed immediately, and stored at -80°C until analysis. The PRAQ questionnaire was administered in a private setting to ensure confidentiality and reduce response bias. All data were recorded and verified before statistical analysis.

Statistical analysis

For statistical analysis, cortisol levels were divided into high and normal, while the PRAG scores were divided into high (≥30) and low-moderate (<30). The associations between dichotomized cortisol levels and PRAQ scores with the occurrence of preeclampsia were examined using the Chi-square test. To quantify the magnitude of the association, the crude odds ratio (OR) and 95% confidence interval (95%CI) were calculated based on the 2×2 contingency table. The OR and its confidence interval were derived using the logarithmic (log-transformed) method. The Spearman's correlation was used to determine the correlation between cortisol levels and PRAG

scores. A *p*-value of <0.05 was considered statistically significant. Data were analyzed using SPSS Statistics version 26 (IBM, New York, USA).

Results

Participant characteristics

A total of 66 third-trimester pregnant women met the eligibility criteria and were enrolled, consisting of 33 patients with preeclampsia and 33 with normal pregnancies. The mean age of participants was 33.3 years. Most participants were married (96.9%), and the majority held secondary or higher education. The two groups were comparable with respect to age, educational level, occupation, parity, and gestational age ($p>0.05$ for all comparisons), confirming group homogeneity. Detailed demographic and clinical characteristics are presented in **Table 1**.

Table 1. Characteristics of research subjects included in this study (n=66)

Characteristic	Preeclampsia (n=33)	Normal pregnancy (n=33)	*p*-value
Age (years), mean±SD	34.2±5.9	32.5±6.4	0.267
Education			
High school	10 (30.3%)	12 (36.4%)	0.548
Bachelor degree	18 (54.5%)	15 (45.5%)	
Master degree	5 (15.2%)	6 (18.2%)	
Occupation			
Housewife	14 (42.4%)	16 (48.5%)	0.394
Civil servant	11 (33.3%)	8 (24.2%)	
Private employee	8 (24.2%)	9 (27.3%)	
Marital status			
Married	32 (97.0%)	32 (97.0%)	1.000
Divorced	1 (3.0%)	1 (3.0%)	
Parity			
Primigravida	13 (39.4%)	14 (42.4%)	0.803
Multigravida	20 (60.6%)	19 (57.6%)	
Gestational age (weeks), mean±SD	36.4 ± 1.8	36.2 ± 1.9	0.665

Association between serum cortisol levels and preeclampsia

A significant association was observed between maternal cortisol levels and the occurrence of preeclampsia (**Table 2**). The majority of women with preeclampsia had high cortisol levels (30/33; 90.9%), whereas only 8 out of 33 women with normal pregnancies (24.2%) had high cortisol levels. Women with high serum cortisol levels had a substantially greater likelihood of developing preeclampsia compared with those with normal cortisol levels, with an OR of 34.00 (95%CI: 4.93–234.46) (**Table 2**). These findings indicate that elevated cortisol was strongly associated with preeclampsia and may serve as an important biological marker for identifying women at increased risk.

Table 2. Association between cortisol levels and preeclampsia occurrence

Cortisol level	Preeclampsia	Normal pregnancy	OR (95% CI)
High	30 (90.9%)	8 (24.2%)	34.00 (4.93–234.46)
Normal	3 (9.1%)	25 (75.8%)	

Association between pregnancy-related anxiety and preeclampsia

A clear association was observed between pregnancy-related anxiety and the occurrence of preeclampsia (**Table 3**). The majority of women with preeclampsia had high PRAQ scores, accounting for 27 out of 33 cases (81.8%). Conversely, low–moderate PRAQ scores were more common among normotensive pregnant women (27/33; 81.8%) compared with those with preeclampsia (6/33; 18.2%). Women with high pregnancy-related anxiety demonstrated a markedly increased likelihood of developing preeclampsia. The calculated odds ratio was 16.71 (95%CI: 4.95–56.39) (**Table 3**), indicating that pregnant women with elevated PRAQ scores were more than 16 times more likely to experience preeclampsia compared with those reporting low to moderate anxiety. This strong association highlights the potential role of psychological stress as

an independent risk factor for preeclampsia and underscores the relevance of anxiety screening during routine antenatal care.

Table 3. Association between Pregnancy Related Anxiety Questionnaire (PRAQ) scores and preeclampsia occurrence

PRAQ score	Preeclampsia	Normal pregnancy	OR (95%CI)
High (≥30)	27 (81.8%)	6 (18.2%)	16.71 (4.95–56.39)
Low-moderate (<30)	6 (18.2%)	27 (81.8%)	

Correlation between serum cortisol levels and pregnancy-related anxiety

Our data indicated no significant correlation was found between serum cortisol concentrations and pregnancy-related anxiety scores in either the preeclampsia or normal pregnancy groups (**Table 4**). Among women with preeclampsia, the correlation was negligible (r=−0.041, p=0.821), indicating that variations in cortisol levels did not correspond meaningfully with changes in PRAQ scores. Similarly, in the normal pregnancy group, a weak but non-significant positive correlation was observed (r=0.278, p=0.117), suggesting only a minimal tendency for higher cortisol levels to accompany higher anxiety scores, though this pattern was not statistically supported.

Table 4. Correlation between cortisol levels and Pregnancy Related Anxiety Questionnaire (PRAQ) scores in the preeclampsia group

Study group	Variable	Mean±SD	Min-Max	Correlation coefficient	p-value
Preeclampsia group	Cortisol (µg/dL)	28.3±5.9	19.2–41.4	-0.041	0.821
	PRAQ score	34.8±6.2	18–44		
Normal pregnancy	Cortisol (µg/dL)	19.4±4.2	12.8–29.2	0.278	0.117
	PRAQ score	24.2±5.7	14–35		

Discussion

This study demonstrated that both elevated serum cortisol levels and high pregnancy-related anxiety were independently associated with the occurrence of preeclampsia among third-trimester pregnant women. However, no significant correlation was found between cortisol levels and anxiety scores in either preeclamptic or normotensive pregnancies. These findings suggest that hormonal and psychological stress pathways contribute separately rather than synergistically to the pathophysiology of preeclampsia.

Preeclampsia is increasingly recognized as a multisystem disorder involving placental dysfunction, systemic inflammation, and maladaptive maternal stress responses [1-4]. The present findings corroborate prior work showing excessive activation of the hypothalamic–pituitary–adrenal axis and dysregulated cortisol metabolism in preeclampsia [5-7]. Cortisol may contribute to vascular pathology by inducing vasoconstriction of uteroplacental arteries, impairing endothelial nitric-oxide synthesis, and increasing oxidative stress [8,9]. Elevated cortisol also alters immune tolerance at the maternal–fetal interface, potentiating cytokine imbalance and endothelial damage [10].

The high odds ratio of 34 highlights the magnitude of cortisol's association with preeclampsia risk. Earlier physiological studies established that cortisol rises progressively during normal pregnancy to promote fetal organ maturation [11,12]. However, exaggerated increases, as observed in preeclamptic women, reflect maladaptive neuroendocrine responses linked to placental hypoxia, impaired 11β-hydroxysteroid dehydrogenase type 2 (11β-HSD2) activity, and elevated placental corticotropin-releasing hormone [13-15]. Inhibition of 11β-HSD2 exposes the fetus to maternal glucocorticoids, thereby restricting growth and compounding perinatal morbidity [16].

Our findings are consistent with reports from both low- and high-income settings describing hypercortisolemia and altered glucocorticoid receptor expression in preeclampsia [17,18]. They also align with mechanistic models demonstrating that chronic stress-induced hypothalamic–pituitary–adrenal axis activation disrupts angiogenic balance through increased soluble fms-like tyrosine kinase-1 (sFlt-1) and decreased placental growth factor (PlGF) [19,20]. Together, these

indicate that cortisol elevation is not merely a by-product of disease but a plausible pathogenic mediator.

Psychological distress and anxiety during pregnancy have long been implicated in adverse obstetric outcomes, including hypertensive disorders, preterm birth, and low birth weight [21]. The present study supported these observations, revealing that women with high pregnancy-specific anxiety had nearly 17-fold greater odds of developing preeclampsia than those with lower anxiety levels. This association mirrors the previous meta-analysis showing a 1.5–2-fold increased risk of preeclampsia in women exposed to high stress [22].

Pregnancy-related anxiety may exacerbate disease risk through sympathetic overactivity, heightened catecholamine release, and pro-inflammatory cytokine production, which collectively impair placental perfusion and vascular reactivity [23-25]. Neuroimaging studies have shown that chronic anxiety modifies limbic–hypothalamic signaling, further stimulating cortisol secretion and inflammatory pathways [26]. The PRAQ specifically quantifies anxieties concerning fetal health, childbirth, and self-perception, offering higher predictive accuracy for obstetric complications than general anxiety scales [27]. Furthermore, culturally mediated stressors—such as socioreligious expectations, limited social support, and financial strain are highly relevant in settings like Aceh, Indonesia, where psychosocial stress may interact with physiological vulnerability [28]. This contextual dimension highlights the importance of region-specific assessment tools and holistic antenatal care models integrating psychological support.

Notably, the absence of a direct correlation between cortisol levels and PRAQ scores challenges the assumption of a simple linear psychoneuroendocrine relationship. Several explanations are plausible. First, cortisol elevation in preeclampsia may arise primarily from placental and endothelial pathology rather than subjective anxiety [3,11]. Second, inter-individual differences in glucocorticoid receptor sensitivity, cortisol-binding globulin, and enzymatic conversion (11β-HSD1/2 balance) could decouple psychological perception from biochemical output [12,16]. Third, inflammatory mediators and placental corticotropin-releasing hormone act as independent stimulants of the hypothalamic–pituitary–adrenal axis, further masking any anxiety–cortisol correlation [14,19]. Previous studies in similar populations also reported weak or absent associations between self-reported stress and serum cortisol, emphasizing that biochemical and psychometric stress markers may capture distinct biological dimensions [17,24]. This divergence reinforces the need to interpret cortisol and anxiety as complementary, not interchangeable, indicators of maternal adaptation to pregnancy stress.

From a clinical standpoint, these results support the implementation of dual-screening models that combine physiological (cortisol) and psychological (PRAQ) assessments within routine antenatal care. Early identification of women with elevated cortisol or anxiety may enable timely interventions—such as stress-reduction counseling, cognitive-behavioral therapy, or pharmacologic modulation of stress responses—to mitigate endothelial dysfunction and hypertensive complications [29]. At a research level, our findings advocate for multidisciplinary approaches uniting obstetrics, endocrinology, and behavioral science. Longitudinal studies with repeated cortisol measurements across trimesters, incorporation of salivary or hair cortisol as chronic stress biomarkers, and integration of inflammatory and angiogenic markers could clarify causal pathways. Intervention trials targeting both stress perception and hypothalamic pituitary adrenal axis modulation may further validate this dual-pathway hypothesis.

The major strength of this study lies in its comprehensive design, integrating hormonal, psychological, and clinical dimensions across multiple tertiary centers in Aceh. Standardized sampling times (06:00–12:00 h) minimized diurnal variation in cortisol, and use of the validated Indonesian PRAQ enhanced measurement reliability. Limitations include the cross-sectional case–control design, which restricts causal inference; single-time cortisol measurement; and moderate sample size, which may limit subgroup analyses. Despite these constraints, the findings provide a robust foundation for future longitudinal and interventional research.

Conclusion

This study provides evidence that both elevated serum cortisol levels and high pregnancy-related anxiety are independently associated with preeclampsia, yet no direct relationship exists between these two stress markers. The findings support the role of combined hormonal and psychological

screening as an innovative strategy for early identification of preeclampsia risk. Integrating such dual assessment into antenatal care could contribute to reducing maternal and neonatal morbidity in resource-limited settings such as Aceh, Indonesia.

Ethics approval

Ethical approval was obtained from the Health Research Ethics Committee of Dr Zainoel Abidin Hospital (No. 112/ETIK-RSUDZA/2025).

Acknowledgments

We would like to thank healthcare workers from Dr. Zainoel Abidin Hospital, Rumah Sakit Ibu dan Anak, and Meuraxa General Hospital who were involved in this study.

Competing interests

All the authors declare that there are no conflicts of interest.

Funding

This study received no external funding.

Underlying data

Derived data supporting the findings of this study are available from the corresponding author on request.

Declaration of artificial intelligence use

We hereby confirm that no artificial intelligence (AI) tools or methodologies were utilized at any stage of this study, including during data collection, analysis, visualization, or manuscript preparation. All work presented in this study was conducted manually by the authors without the assistance of AI-based tools or systems.

How to cite

Hezron TM, Dewi TP, Septivera Y, *et al*. Cortisol and pregnancy-related anxiety in relation to preeclampsia among third-trimester pregnant women: A case–control study from Aceh, Indonesia. Narra J 2025; 5 (3): e2985 - http://doi.org/10.52225/narra.v5i3.2985.

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