DOI: https://doi.org/10.54393/pjhs.v7i10.3985

PAKISTAN JOURNAL OF HEALTH SCIENCES
(LAHORE)
https://thejas.com.pk/index.php/pjhs
ISSN (E): 2790-9352, (P): 2790-9344
Volume 7, Issue 10 (October 2026)



OPEN ACCESS

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Original Article

Comparison of Serum Calcium-Magnesium Ratio in Women with and without Preeclampsia

Abida Rehman[1*], Saima Zulfiqar[1], Shazia Zulfiqar[2], Sadia Zulfiqar[1], Sana Ali[3] and Farhana[1]

[1]Department of Obstetrics and Gynecology, Sheikh Zayed Hospital, Rahim Yar Khan, Pakistan
[2]Department of Obstetrics and Gynecology, Lahore Medical and Dental College, Lahore, Pakistan
[3]Department of Obstetrics and Gynecology, Ghurki Trust Teaching Hospital, Lahore, Pakistan

ARTICLE INFO

Keywords:
Preeclampsia, Serum Calcium, Serum Magnesium, Calcium to Magnesium Ratio, Pregnancy-Induced Hypertension

How to Cite:
Rehman, A., Zulfiqar, S., Zulfiqar, S., Zulfiqar, S., Ali, S., & Farhana, . (2026). Comparison of Serum Calcium-Magnesium Ratio in Women with and without Preeclampsia: Serum Calcium-Magnesium Ratio in Preeclamptic Women. Pakistan Journal of Health Sciences, 7(10), 136-141. https://doi.org/10.54393/pjhs.v7i10.3985

***Corresponding Author:**
Abida Rehman
Department of Obstetrics and Gynecology, Sheikh Zayed Hospital, Rahim Yar Khan, Pakistan
drabidarehman@yahoo.com

Received Date: 17th February, 2026
1st Revision Received: 23rd April, 2026
2nd Revision Received: 12th May, 2026
Acceptance Date:1st June, 2026
Published Date: 31st October, 2026

ABSTRACT

Preeclampsia remains a major cause of maternal and perinatal morbidity, and altered calcium and magnesium balance may contribute to its pathophysiology. **Objective:** To compare serum calcium, serum magnesium, and the calcium-to-magnesium ratio between women with preeclampsia and normotensive pregnant women. **Methods:** A comparative cross-sectional study was conducted in the Department of Obstetrics and Gynecology at a tertiary care hospital from July to December 2025. Using non-probability consecutive sampling, 52 women at or beyond 24 weeks' gestation were enrolled. Cases were diagnosed with preeclampsia, and controls were normotensive with negative urine protein. Serum calcium and magnesium were measured to calculate the calcium-to-magnesium ratio. **Results:** Fifty-two pregnant women were analyzed, including 26 with preeclampsia and 26 normotensive controls. Maternal age was similar between groups (28.96 ± 2.76 vs 29.54 ± 3.00 years, p=0.474), and gestational age did not differ (33.04 ± 2.16 vs 32.92 ± 3.01 weeks, p=0.874). Body mass index was higher in preeclampsia (28.57 ± 4.86 vs 24.27 ± 4.38 kg/m^2, p=0.002). Systolic and diastolic pressures were higher in preeclampsia (p<0.001 each). Serum calcium, magnesium, and the calcium to magnesium ratio were lower in preeclampsia (p<0.001, p=0.015, and p=0.002), supporting altered mineral balance in affected pregnancies. **Conclusion:** Women with preeclampsia showed lower serum calcium, serum magnesium, and a reduced calcium to magnesium ratio compared with controls. Consequently, a low serum calcium to magnesium ratio could be a risk factor for preeclampsia.

INTRODUCTION

Preeclampsia is a hypertensive disorder of pregnancy that typically presents after 20 weeks of gestation with new-onset hypertension accompanied by proteinuria and or maternal end-organ dysfunction [1]. It remains a major contributor to maternal and perinatal morbidity and mortality, with population estimates commonly ranging between 2% and 5% of pregnancies worldwide [2]. A recent systematic review and meta-analysis pooling more than two million pregnancies reported a global prevalence of preeclampsia of 4.43% (95% confidence interval 3.73% to 5.20%) [3]. A multicenter study (2020–2022) reported 3% preeclampsia among obstetric admissions, while national Pakistani summaries cited in a Lahore case control study reported an approximately 2.4% incidence [4, 5]. Risk is increased by pre-existing hypertension, diabetes mellitus, chronic kidney disease, obesity, and autoimmune disorders, and is exacerbated by delayed detection and limited specialist care in resource-limited settings [6]. Preeclampsia is multifactorial, driven by abnormal placentation, oxidative stress, and endothelial dysfunction; however, routine use of angiogenic biomarkers is limited by cost and laboratory constraints,

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DOI: https://doi.org/10.54393/pjhs.v7i10.3985

supporting evaluation of accessible biochemical markers [7]. Calcium and magnesium are mechanistically relevant because calcium influences vascular smooth muscle contractility and renin release, whereas magnesium acts as a physiological calcium antagonist with vasodilatory and anti-inflammatory effects [8]. In low-calcium-intake populations, calcium supplementation has been associated with an approximate halving of preeclampsia risk. A systematic review and meta-analysis including 10,482 pregnancies reported significantly lower serum calcium in preeclampsia compared with normotensive pregnancy (weighted mean difference −0.807 mg/dL, 95% confidence interval −0.911 to −0.703), alongside lower serum magnesium (weighted mean difference −0.215 mg/dL, 95% confidence interval −0.281 to −0.148). However, composite indices may better reflect the shared biology of these ions than single measures. In a study, the serum calcium to magnesium ratio was significantly lower in women with preeclampsia than in controls (1.98 vs 2.60; p<0.001), with a proposed cut-off of 2.36 yielding modest discrimination [9]. Conversely, other datasets have shown no meaningful difference in the ratio overall, while suggesting a potential gradient with severity, with lower ratios observed in severe versus mild preeclampsia [10].

Despite increasing evidence on the role of calcium and magnesium in the pathophysiology of preeclampsia, the findings from previous studies remained inconsistent, particularly regarding the predictive value of the calcium-to-magnesium ratio. Therefore, clarification of the relationship between mineral homeostasis and preeclampsia remains necessary. In addition, most studies have been conducted in diverse populations with limited applicability to local settings, and accessible biochemical markers suitable for routine clinical use remain inadequately explored. Therefore, this study aimed to compare the serum calcium to magnesium ratio in women with preeclampsia and normotensive pregnant controls.

METHODS

A comparative cross-sectional study was conducted in the Department of Obstetrics and Gynecology, Sheikh Zayed Hospital, Rahim Yar Khan, from July to December 2025. Participants were enrolled through non-probability consecutive sampling after taking informed written consent, and ethical approval (227/IRB/SZMC/SZH dated 19th July 2025) was obtained from the institutional review board. The sample size was calculated using a 95% confidence level and 90% statistical power, based on previously reported calcium-to-magnesium ratio estimates of 3.36 ± 0.60 among women with preeclampsia and 3.83 ± 0.41 among normotensive pregnant women [11], resulting in a required sample of 52 participants (26 per group). Consecutive sampling was selected to facilitate recruitment of eligible participants during the study period; however, its non-probability nature may have introduced selection bias. Eligible participants were pregnant women at or beyond 24 weeks of gestation. The cases were women diagnosed with preeclampsia, and the controls were normotensive pregnant women with a negative urine dipstick for protein. Exclusion criteria included chronic hypertension, chronic medical illnesses that could confound serum calcium or magnesium assessment, use of diuretics or calcium or magnesium supplements, renal disorders or other conditions affecting calcium or magnesium metabolism, congenital fetal anomalies on anomaly scan, multiple gestation, diabetes mellitus, autoimmune disease, thyroid or parathyroid disease, and hypertension developing before 24 weeks of gestation. Baseline demographic and clinical information, including maternal age, gestational age, gravidity, parity, and body mass index, was collected at enrolment using a specifically designed data collection form. The BMI was calculated using the standard method, weight in kilograms divided by height in meters squared (kg/m^2). For categorical analysis, standard adult cutoffs were used: underweight <18.5 kg/m^2, normal weight 18.5 to 24.9 kg/m^2, overweight 25.0 to 29.9 kg/m^2, and obesity ≥30.0 kg/m^2. Because the study was conducted during pregnancy, body mass index was considered a baseline anthropometric measure rather than a measure of pre-pregnancy adiposity. Blood pressure was measured using a calibrated sphygmomanometer with an appropriately sized cuff. Measurements were obtained after a rest period with the participant seated and the arm supported at heart level. Urinary protein was assessed using urine dipstick testing performed as part of routine obstetric evaluation. Group allocation was performed at enrolment using standard diagnostic criteria. The exposure category was labelled as "Diagnosed with Preeclampsia" or "Control (Without Preeclampsia)" based on clinical assessment. Preeclampsia was defined as new-onset hypertension arising at or after 20 weeks of gestation accompanied by proteinuria, consistent with international guidance. Hypertension was defined as systolic blood pressure ≥140 mmHg and or diastolic blood pressure ≥90 mmHg on at least two measurements, separated by an appropriate interval, using a standardized technique. Controls were defined as pregnant women without preeclampsia, with systolic blood pressure <140 mmHg and diastolic blood pressure <90 mmHg, and without documented proteinuria at the time of enrolment. Venous blood sampling was performed at enrolment under aseptic technique. Samples were collected before administration of any acute intravenous mineral preparations if such treatment was planned clinically. Blood was processed according to laboratory standard

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operating procedures, including serum separation by centrifugation. Serum calcium and serum magnesium were measured in mg/dl using an automated chemistry analyzer with commercially available reagent kits, following manufacturer instructions. The primary biochemical outcome, the calcium to magnesium ratio, was calculated by dividing serum calcium by serum magnesium using the same units, and the derived ratio was recorded in the form.

Data were analyzed in IBM SPSS Statistics version 26.0. Continuous data were summarized as mean ± standard deviation and compared using the independent-samples t test, while categorical data were reported as frequency (%) and compared using the Pearson chi-square. A two-sided p-value <0.05 was considered statistically significant.

RESULTS

A total of 52 pregnant women were analyzed, including 26 diagnosed with preeclampsia and 26 controls. Mean maternal age did not differ between groups (28.96 ± 2.76 vs 29.54 ± 3.00 years, p=0.474), and gestational age was comparable (33.04 ± 2.16 vs 32.92 ± 3.01 weeks, p=0.874). In contrast, body mass index was significantly higher in the preeclampsia group (28.57 ± 4.86 vs 24.27 ± 4.38 kg/m^2, p=0.002)(Table 1).

Table 1: Baseline Demographic and Obstetric Characteristics of Women with and without Preeclampsia(n=52)

Variables	Preeclampsia, Mean ± SD, (n=26)	Control, Mean ± SD, (n=26)	Mean Difference (95% CI)	p-value
Maternal Age (Years)	28.96 ± 2.76	29.54 ± 3.00	−0.58 (95% CI −2.18 to 1.03)	0.474
Gestational Age (Weeks)	33.04 ± 2.16	32.92 ± 3.01	0.12 (95% CI −1.34 to 1.57)	0.874
Body Mass Index (kg/m^2)	28.57 ± 4.86	24.27 ± 4.38	4.31 (95% CI 1.73 to 6.88)	0.002

Independent-samples t tests were applied with 95% confidence intervals

Gravidity distribution was similar between groups (1–2 pregnancies: 53.8% vs 61.5%, p=0.575). Parity category also did not show a statistically significant difference (0–2 live births: 80.8% vs 61.5%, p=0.126). The distribution of BMI categories did not differ significantly between the two groups (p=0.058)(Table 2).

Table 2: Gravidity, Parity, and Body Mass Index Categories by Group(n=52)

Variables	Preeclampsia (n=26), n (%)	Control (n=26), n (%)	Odds Ratio (95% CI)	p-value
Gravidity Category				
1–2	14 (53.8%)	16 (61.5%)	1.37 (0.46–4.13)	0.575
≥ 3	12 (46.2%)	10 (38.5%)		
Parity Category				
0–2	21 (80.8%)	16 (61.5%)	0.38 (0.11–1.34)	0.126
≥ 3	5 (19.2%)	10 (38.5%)		
Body Mass Index Category				
<18.5	0 (0.0%)	2 (7.7%)	–	0.058
18.5–24.9	7 (26.9%)	14 (53.8%)	–	
25.0–29.9	11 (42.3%)	7 (26.9%)	–	
≥30.0	8 (30.8%)	3 (11.5%)	–	

Systolic and diastolic blood pressure values were significantly higher in the preeclampsia group. Mean systolic blood pressure was 151.69 ± 10.57 mmHg in preeclampsia versus 119.69 ± 9.71 mmHg in controls (p<0.001). Mean diastolic blood pressure was 99.50 ± 5.81 mmHg versus 74.50 ± 6.31 mmHg (p<0.001). Serum calcium, serum magnesium, and the calcium-to-magnesium ratio were all significantly lower in women with preeclampsia. Serum calcium was 8.10 ± 0.56 mg/dL in preeclampsia compared with 9.54 ± 0.61 mg/dL in controls (p<0.001). Serum magnesium was 1.89 ± 0.27 mg/dL versus 2.06 ± 0.22 mg/dL (p=0.015). The calcium to magnesium ratio was 3.25 ± 0.54 in preeclampsia compared with 3.68 ± 0.41 in controls (p=0.002)(Table 3).

Table 3: Clinical and Biochemical Comparisons Between Groups(n=52)

Variables	Preeclampsia (n=26) Mean ± SD	Control (n=26) Mean ± SD	Mean difference (95% CI)	p-value
Systolic Blood Pressure (mmHg)	151.69 ± 10.57	119.69 ± 9.71	32.00 (26.35 to 37.65)	<0.001
Diastolic Blood Pressure (mmHg)	99.50 ± 5.81	74.50 ± 6.31	25.00 (21.62 to 28.38)	<0.001
Serum Calcium (mg/dL)	8.10 ± 0.56	9.54 ± 0.61	−1.43 (−1.76 to −1.10)	<0.001
Serum Magnesium (mg/dL)	1.89 ± 0.27	2.06 ± 0.22	−0.17 (−0.30 to −0.03)	0.015
Calcium to Magnesium Ratio	3.25 ± 0.54	3.68 ± 0.41	−0.43 (−0.70 to −0.16)	0.002

Independent-samples t tests were applied with 95% confidence intervals

DISCUSSION

The present study evaluated serum calcium, serum magnesium, and the calcium to magnesium ratio in pregnant women, comparing with preeclampsia to normotensive controls. Mean maternal age and gestational age were comparable between the groups in the present study, with mean age 28.96 ± 2.76 years in preeclampsia versus 29.54 ± 3.00 years in controls (p=0.474), and mean gestational age 33.04 ± 2.16 versus 32.92 ± 3.01 weeks (p=0.874). Similar baseline comparability for age

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DOI: https://doi.org/10.54393/pjhs.v7i10.3985

and gestational age has been reported in several studies, which reported closely matched ages and gestational ages. Such comparability strengthens the interpretation that observed biochemical differences are less likely to be driven by these two variables alone [11, 12]. Body mass index was significantly higher in preeclampsia in the present study (28.57 ± 4.86 vs 24.27 ± 4.38 kg/m^2, p=0.002), although the distribution of categorical BMI groups did not differ significantly (p=0.058). A similar association between higher body mass index and preeclampsia has been repeatedly observed in prior datasets, including Sah and Goel, where body mass index was higher among cases (27.17 ± 4.58 vs 24.44 ± 3.63 kg/m^2) [13]. Given established links between adiposity, systemic inflammation, endothelial dysfunction, and preeclampsia risk, body mass index also represents a plausible confounder. Blood pressure differences in the present study were substantial and consistent with preeclampsia physiology, with systolic blood pressure 151.69 ± 10.57 mmHg versus 119.69 ± 9.71 mmHg, and diastolic blood pressure 99.50 ± 5.81 mmHg versus 74.50 ± 6.31 mmHg, both p<0.001. These values fall within the range reported by comparative studies, including Mittal *et al.* (145.32 ± 6.66 and 96.8 ± 5.00 in preeclampsia), Sethi *et al.* (154.6 ± 13.43 and 99.0 ± 8.63), Aslam *et al.* (155.890 ± 7.799 and 104.380 ± 5.729), and Okoror *et al.* reporting very high blood pressures in cases [11, 14, 15]. Serum calcium was significantly lower in preeclampsia in the present study (8.10 ± 0.56 mg/dL) than in controls (9.54 ± 0.61 mg/dL), with a mean difference of −1.43 mg/dL (p<0.001). This finding is consistent with previous studies, including Aslam *et al.* who reported calcium levels of 9.08 ± 0.63 mg/dL in controls, 7.91 ± 0.64 mg/dL in women with mild preeclampsia, and 7.67 ± 0.48 mg/dL in women with severe preeclampsia [16]. The concordance supports case classification validity and provides a biological context for investigating mineral indices, which may relate to vascular tone and endothelial function. Serum calcium was significantly lower in preeclampsia in the present study (8.10 ± 0.56 mg/dL) than in controls (9.54 ± 0.61 mg/dL), with a mean difference of −1.43 mg/dL (p<0.001). This reduction is consistent with several comparative studies reporting lower calcium among preeclamptic women, including Aslam *et al.* (9.08 ± 0.63 vs 7.67 ± 0.48), Aslam *et al.* (7.90 ± 0.29 vs 9.22 ± 0.59), Sethi *et al.* using ionic calcium (3.34 ± 1.35 vs 4.37 ± 0.74) and the longitudinal data of Rifat and Al-Rawi showing lower calcium in women who developed preeclampsia later in gestation [15-18]. Serum magnesium was lower in preeclampsia in the present study (1.89 ± 0.27 mg/dL) than in controls (2.06 ± 0.22 mg/dL, p=0.015), aligning with several reports showing hypomagnesemia in preeclampsia and, in some datasets, a severity related decline, including Aslam *et al.* (2.12 ± 0.22 in controls vs 1.58 ± 0.42 in mild and 1.26 ± 0.44 in severe), Aslam *et al.* (1.770 ± 0.228 vs 1.957 ± 0.13394), Sethi *et al.* (1.68 ± 0.41 vs 1.94 ± 0.42), Mittal *et al.* (1.99 ± 0.13 vs 2.03 ± 0.13), and the longitudinal findings of Rifat and Al-Rawi at later gestation [15-17]. However, magnesium shows greater heterogeneity than calcium, with null differences reported by Okoror *et al.* and Sah and Goel, and higher magnesium variation related to assay methods, gestational timing, supplementation, and renal or hemoconcentration effects [11, 13]. Despite this, pooled evidence supports an overall reduction in preeclampsia, with Eslamzadeh *et al.* demonstrating a significant pooled difference across 64 studies. Low calcium in preeclampsia may increase vascular smooth muscle tone and peripheral resistance [18]. Reduced extracellular calcium can promote parathyroid hormone release and intracellular calcium influx, favouring vasoconstriction. Magnesium acts as a physiological calcium antagonist and supports endothelial function; reduced magnesium may further facilitate vasospasm. The calcium to magnesium ratio was significantly lower in preeclampsia in the present study (3.25 ± 0.54) than in controls (3.68 ± 0.41, p=0.002), consistent with Okoror *et al.* (3.36 ± 0.60 vs 3.83 ± 0.41, p=0.001) reported a reduced ratio [11, 12]. This composite index may reflect concurrent calcium depletion and magnesium alteration, thereby summarizing the mineral balance relevant to vascular tone. However, because the ratio is derived from two correlated measures, it is susceptible to confounding and propagation of measurement error. Okoror *et al.* showed attenuation of the ratio association after adjustment, while hypocalcemia remained independently associated, indicating that the ratio may partly mirror calcium status and baseline covariates such as adiposity [11]. Diagnostic performance has varied across reports, with moderate discrimination in Onyegbule *et al.* and higher discrimination in Torres-Cepeda *et al.* and differing cut-offs, suggesting that thresholds are unlikely to be transferable without local calibration [19, 20]. Accordingly, the present finding supports the ratio as a supportive marker, but larger studies with multivariable modelling and external validation are required before clinical implementation.

This study has several limitations. The relatively small sample size may have limited statistical power and the precision of effect estimates. The single-center design and use of non-probability consecutive sampling may have introduced selection bias and may limit the generalizability of the findings to other populations and settings. BMI was assessed at enrolment during pregnancy and therefore may not accurately reflect pre-pregnancy adiposity. Dietary calcium and magnesium intake, micronutrient supplementation, and serum albumin were not assessed

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DOI: https://doi.org/10.54393/pjhs.v7i10.3985

and may have influenced circulating mineral concentrations, particularly total serum calcium. BMI differed significantly between groups; however, multivariable adjustment was not performed because of the limited sample size, so residual confounding cannot be excluded. Multiple statistical comparisons were performed without adjustment for multiplicity, which may have increased the possibility of type I error. Furthermore, the calcium-to-magnesium ratio is a derived measure based on two correlated biochemical variables, and formal assessment of statistical dependence and measurement-error propagation was not performed. Finally, diagnostic accuracy was not assessed, and therefore no sensitivity, specificity, AUC, or clinically applicable cut-off value can be established from the present data. Larger, multicenter studies incorporating multivariable modelling, ROC analysis, and external validation are needed to determine the clinical relevance of the calcium-to-magnesium ratio.

CONCLUSIONS

Women with preeclampsia had lower serum calcium, serum magnesium, and calcium-to-magnesium ratio compared with normotensive pregnant women. These findings indicate an association between altered mineral status and preeclampsia; however, causality and diagnostic or predictive utility of the ratio cannot be established from this cross-sectional study.

Authors' Contribution

Conceptualization: F
Methodology: AR, SZ
Formal analysis: SZ, SZ
Writing and Drafting: AR, SZ, SZ, F
Review and Editing: AR, SZ, SZ, SZ, F

All authors approved the final manuscript and take responsibility for the integrity of the work

Conflict of Interest

All the authors declare no conflict of interest.

Source of Funding

The authors received no financial support for the research, authorship, and/or publication of this article.

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