## ORIGINAL ARTICLE

# ROLE OF RANDOM URINARY PROTEIN TO CREATININE RATIO IN MILD PREECLAMPSIA

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## ABSTRACT

**Introduction:** Preeclampsia is one of the important members of deadly triad, along with hemorrhage and infection which contributes for the maternal morbidity and mortality rates. Proteinuria is an important diagnostic component of preeclampsia.
**Aim**: To compared the results of s random urine protein-creatinine (P/C) ratio with 24-hour urine protein excretion in women with mild preeclampsia.
**Methodology:** 100 pregnant women with mild preeclampsia as cases and age matched 100 normal healthy pregnant women as controls were studied for proteinuria. Urine P/C ratio was determined in a random sample, and the amount of protein excretion was measured in 24-hour urine collected on the subsequent day and compared between cases and age matched controls of age group 18-35 years using unpaired two-tailed Student 't' test. The correlation between the spot P/C ratio and 24-hour urine protein excretion was assessed. The receiver operating characteristic curve analysis was used to determine the best discriminator values of the spot urine P/C ratios for preeclampsia (proteinuria ≥ 300 mg/24 h). All statistical analyses were performed using GRAPH PAD PRISM version 5.00 software.
**Results:** There was a strong correlation between the spot P/C ratio and 24-hour urine protein excretion ($r = 0.94$; $P < .0001$). ROC curve reveals area 0.95. The optimal spot P/C ratio cutoff point was 0.3 for 300 mg/24 h of protein excretion (preeclampsia), with a sensitivity, specificity, positive predictive value, and negative predictive value of 94%, 94%, 94.4%, and 96.8%, respectively.
**Conclusion:** We found that there is a significant correlation between the spot urine P/C ratio and 24-hour urine protein excretion in women with preeclampsia. Urine P/C ratio could be used for diagnosis as well as screening of mild preeclampsia.

**Keywords:** Preeclampsia, Proteinuria, Protein to creatinine (P/C) ratio

## INTRODUCTION

Hypertensive disorders accounts for 5-10% of all pregnancies, and considering their complications, they are among the major causes of maternal morbidity and mortality. [1, 2] Preeclampsia, the most prevalent hypertensive disorders of pregnancy, is defined as a systolic blood pressure level of 140 mm Hg or higher or a diastolic blood pressure level of 90 mm Hg or higher that occurs after 20 weeks of gestation with proteinuria (≥300 mg/24 hrs). Preeclampsia can be mild or severe. In severe preeclampsia, patients have renal failure with high serum creatinine, more edema, BP>160/110 mmhg and 3+ or more protein on dipstick, where diagnosis is very clear, patients are immediately started on treatment. Practically no time permits for 24 hrs urine collection and first priority is treatment.

In mild preeclampsia BP is >140/90 mmhg but <160/110 mmhg, normal creatinine and proteinuria > 0.3 gm/24 hrs. So that in mild preeclampsia for diagnosis 24 hrs urine is must along with BP > 140/90 mmhg. But the difficulties in 24 hrs urinary protein estimation are very well known. This test is unreliable in one third cases. [3] It is time consuming and also prolongs the patient's hospital stays. Detection of proteinuria in a single random urine sample can be an alternative to the timed urine collections in pre-eclampsia. [4] So that random urinary proteins to creatinine ratio measurement provide a good and fast estimation total 24 hrs proteinuria in hospitalized pregnant woman and can replace the time-consuming 24 hrs urine colletions.[5, 6] To overcome these difficulties single random urinary protein to creatinine ratio is good alternative. So we planned to study this ratio in mild preeclampsia and to determine whether it can replace 24 hrs urinary protein estimation. Thus early diagnosis and treatment of preeclampsia is possible and its progression to severe stage and complication can be prevented.

## METHODOLOGY

The present study has been carried out in Indira Gandhi Government Medical College & Mayo Hospital, Nagpur. Total study carries out from November 2011 - April 2015. The study protocol was approved by the Institutional Ethical Committee. Informed written consent was obtained from all the study subjects enrolled in the study. Study sample was included total of 200 individuals; 100 diagnosed mild preeclamptic patients (Cases) admitted in ANC ward in this institute and 100 age matched healthy and apparently normal pregnant women (controls) were also selected for study.

The cases and controls were in the age group of 18-35 years.

**Exclusion criteria for cases and controls:** a known kidney disease, heavy exercise (more than 1 hour of vigorous exercise on the day of urine collection), bacteriuria, Urinary tract infections, bed rest longer than 24 hours, and gestational diabetes mellitus.

24 hrs urine protein and random urinary protein to creatinine ratio: [4, 7] Random urine sample for determining urinary protein and creatinine excretion collected in a sterile plain bulb during the daytime before the start of the 24-h urine collections**.** Patients were instructed to separate the labia while collecting the urine sample. The collected samples were analyzed immediately for urine protein and urine creatinine. Urine protein estimation was done with the kit based on Pyrogallol Red Method (End Point). Urine creatinine estimation was done with the kit based on Initial Rate Modified Jaffe's Method. The estimation was done on TRANSASIA ERBA CHEM-5 Plus Semi-Automatic Analyzer. The Urine Protein/Creatinine Ratio was calculated as urinary protein concentration mg/ mg of creatinine**.**

For a 24-hour urine collection, all of the urine that pass over a 24-hour time period must be collected. After collecting random sample, next day in the morning patient instructed to urinate into the toilet after get up in the morning. Afterwards, collect all urine in a special container for the next 24 hours. On day 2 patient had told to urinate into the container when get up in the morning. Then immediately sample was analyzed with the kit based on Pyrogallol Red Method (End Point) on TRANSASIA ERBA CHEM-5 Plus Semi-Automatic Analyzer.

**Statistical Analysis:** Unpaired two-tailed Student't' test was used to assess the significance of the differences in values of the parameters in cases and controls and values were reported as the mean ± SD . Pearson's correlation coefficients were used to analyze linear correlations between variables. Differences were considered statistically significant at a probability value $p < 0.05$. All statistical analyses were performed using GRAPH PAD PRISM version 5.00 software.[8]

## RESULTS

In cases the mean age of distribution was 22.86 ± 2.63 years and gestational age was 34.85 ± 1.03 weeks while in controls the mean age of distribution was 22.95 ± 2.27 years and gestational age was 34.85 ± 1.03. (Table 1)

**Table 1: Comparison of mean age and mean gestational age in cases and controls**

| Parameters | Cases (n=100) | Controls (n=100) | p-value |
|---|---|---|---|
| Mean age ± SD | 22.86 ± 2.625 | 22.95 ± 2.271 | 0.79 |
| Mean Gestational age ± SD | 34.85 ± 1.030 | 34.85 ± 1.028 | 0.97 |
| Systolic blood pressure (mm Hg) | 145.5 ± 2.303 | 105.7 ± 7.536 | <0.0001 |
| Diastolic blood pressure (mm Hg) | 96.32 ± 2.767 | 72.08 ± 4.421 | <0.0001 |
| 24 hrs urine proteins (mg/24 hrs) | 772.6 ± 195.2 | 145.3 ± 26.79 | <0.0001 |
| Urine protein to creatinine ratio (mg/mg) | 0.68 ± 0.24 | 0.22 ± 0.07 | <0.0001 |

On comparing mean age and gestational age of cases and controls by unpaired t test, p value was 0.79 and 0.97 respectively which was statistically non significant. Hence both the groups were comparable.

Systolic blood pressure (SBP) of cases i.e. 145.5 ± 2.30 mm of Hg was higher than that of controls i.e. 105.7 ± 7.54 mm of Hg (Table 1). Statistically, the difference between mean SBP of cases & controls was highly significant ($P < 0.0001$). Cases had diastolic blood pressure (DBP) of 96.32 ± 2.77 mm of Hg which is higher as compared to that of controls which was 72.08 ± 4.42 mm of Hg. (Table 1) The mean DBP of cases & controls has shown a statistically highly significant difference. ($P < 0.001$)

Cases had 24 hrs urine protein level of 772.6 ± 195.2 mg/dl which was significantly higher than levels in controls i.e. 145.3 ± 26.79 mg/L (Table 1). The difference between mean 24 hrs urine proteins levels of cases & controls was highly significant ($P < 0.001$). Urine protein/creatinine ratio of cases which was 0.68 ± 0.24 mg/mg of creatinine was significantly higher than the ratio in controls i.e. 0.22 ± 0.07 mg/mg of creatinine (Table 1). Statistical comparison of the

mean urine protein/creatinine ratio of cases & controls had shown highly significant difference (P < 0.0001).

**Table 2: Correlation of 24 hrs urine protein and random urinary protein to creatinine ratio in cases**

| Parameter | Pearson's Correlation Coefficient (r) | P value |
|---|---|---|
| 24 hrs urine protein Vs Urine protein to creatinine ratio | 0.94 | <0.0001 |

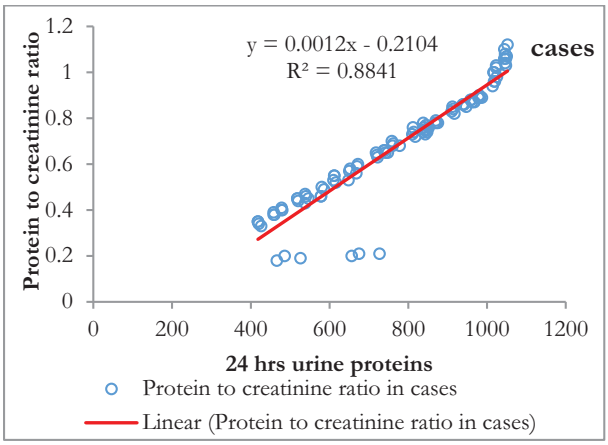


**Figure 1: Correlation of 24 hrs urinary proteins with urinary protein to creatinine ratio**

Correlation study of 24 hrs urine protein with random urinary protein to creatinine ratio in cases indicates that 24 hrs urine proteins was positively correlated with the random urinary protein to creatinine ratio. Pearson's correlation coefficient for this correlation was r = 0.94 and the correlation is statistically highly significant (P < 0.0001). (Table 2 , Fig 1)

The ROC curve for the random urinary protein to creatinine ratio is shown in figure 2. The area under the ROC curve is 0.95 (95% confidence interval: 0.9220 to 0.9852; standard error = 0.016, p value is < 0.0001 i.e. highly significant).The cutoff value of > 0.3 yields a sensitivity of 94% and a specificity of 94%. With the use of this cutoff, there was 6 false–negative and 6 false –positive test results. (Table 3, Fig 2)

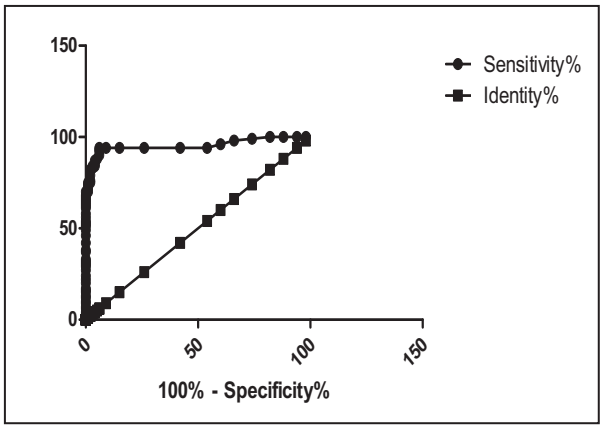


**Figure 2: Random urinary protein/creatinine ratio for the detection of significant proteinuria with the cutoff value 0.3 using ROC analysis**

**Table 3: Result of the random urinary protein/creatinine ratio for the detection of significant proteinuria with the cutoff value 0.3 using ROC analysis**

| | P/C ratio> 0.3 | P/C ratio< 0.3 | Area | Std. Error | 95% confidence interval | P value |
|---|---|---|---|---|---|---|
| Cases | 94 | 6 | 0.95 | 0.01611 | 0.9220 - 0.9852 | < 0.0001 |
| Controls | 6 | 94 | | | | |

## DISCUSSION

Preeclampsia is a syndrome of hypertension in pregnancy, with or without edema and proteinuria. In a number of patients, the clinical appearance is mild, presenting only with small increase in blood pressure or protein in the urine but in other patients severe maternal and fetal complications, such as the HELLP syndrome, eclampsia, preterm delivery, abruptio placenta, intra-uterine fetal death or fetal growth restriction may take place. High blood pressure in pregnant women could cause large placental infarcts and decreased placental growth. It also results in fetal malnutrition, decreased placental perfusion and reduced fetal growth. [9]

One of the ways to diagnose preeclampsia, apart from the blood pressure criteria, is to look for the presence of significant proteinuria. The gold standard for determining protein excretion is the 24 hour urine collection. 24 hours urine collection for estimation of proteins is considered the traditional comparator for quantification of proteinuria in pregnancy. But it has limitations: the urine collection is cumbersome, time consuming, inconvenient, expensive and unreliable in one third cases and potentially misleading if done inaccurately; and collection may not be possible during delivery. Previous studies have demonstrated inadequate collected urine volumes in up to 37% of samples. Waiting for the results of 24-hour urine collection can often delay diagnosis of preeclampsia and may result in prolonged hospital stay for investigations and also increase the cost. [10-12]

The major problem with the 24 hour protein collection is that it is often impractical in the outpatient setting with problems of incomplete collection. Accurately substitution of a spot urine P/C ratio for a 24-

hour urine collection would have significant implications including facilitation of prompt clinical decision making. This would also impact healthcare costs and improve patients' satisfaction with care, too. During the day urinary protein and creatinine excretion rates are fairly constant provided the glomerular filtration rate is constant. Thus a random urinary protein to creatinine ratio in a single voided urine sample can indicate the cumulative excretion during the day since the ratio of two stable rates would cancel out the time factor. [13] Current Australian guidelines advocate use of the spot urine protein-to-creatinine ratio as an alternative to 24 hour urine collection. [14]

Our data also suggested that 24 hrs urine proteins are positively correlated with the random urinary protein to creatinine ratio (Table 2 and Fig 1). Pearson's correlation coefficient for this correlation is $r = 0.94$ and the correlation is statistically highly significant ($P < 0.0001$). This finding is consistent with other studies. [15, 16, 17] However, using the spot P/C ratio of 0.3 as a correlate to the critical value of 300 mg of protein over 24 hours would result in the failure to identify significant proteinuria in approximately 6% of affected patients.

A good correlation between the spot urine P/C ratio and 24-hour protein excretion has been demonstrated in patients with diabetic nephropathy, lupus nephritis, chronic kidney disease, and transplanted kidneys.[3, 18] The National Kidney Foundation guidelines have suggested that spot urine samples should be used to detect and monitor proteinuria in children and adults.[19] These are characterized by excellent accuracy.

Our data suggested that the random urine P/C ratio is a highly accurate test for discriminating between insignificant and significant proteinuria, as demonstrated by an area under the ROC curve of 0.95. (Table 3 and Fig.2) The main concern in clinical use of this test is the false-negative test results, because 6% of patients with preeclampsia may be missed. To obtain the optimal cutoff, we selected the one that while increasing specificity maintains a sensitivity of higher than 90% in order to reduce the possibility of missing the diagnosis of preeclampsia. In our study at 0.3 cutoff both sensitivity and specificity were 94%. A systematic review by Leaños-Miranda A et al (2007) [4] also stated that the protein: creatinine ratio $\geq 0.3$ as an indicator of protein excretion >300 mg/24 h. The sensitivity and specificity were 98.2% and 98.8%, respectively. William's textbook of obstetric has mentioned that urinary protein to creatinine ratio $\geq 0.3$ use for diagnosis of proteinuria. Research in the future should be focused on the evaluation of clinical outcomes and the cost-effectiveness of the use of a random urinary P/C ratio for prediction of significant proteinuria. In addition, studying the test in an outpatient basis should be further considered in order to apply it in ambulatory management of preeclamptic patient.

Alternatives for the diagnosis of proteinuria in pregnancy include urinary dipsticks. The dipstick is inexpensive, easy to use, and provides a rapid result but has been shown to have low sensitivity and specificity for urinary protein excretion over 24 hours. [20-25] Dipsticks poorly quantify 24 hrs proteins. Various studies proved that P: C ratio is more accurate and had better correlation with 24 hrs urine proteins than dipsticks. [16, 24, 26, 27]

The need for a simple reliable screening test to quantitate proteinuria is very important need. So we recommend random urinary protein to creatinine ratio for diagnosis as well as screening purpose, especially in mild preeclamptic patient where for diagnosis proteinuria is must. So that rapid diagnosis and screening of mild preeclamptic patients along with blood pressure is possible and with prompt treatment further progression to severe stage can be prevented. The use of random urinary protein to creatinine ratio as a screening test proved in other studies also. [28-33]

## CONCLUSION

Based on the findings of the present study, we conclude that a random urine P/C ratio predicts the amount of 24-hour urine protein excretion with a highly accuracy. This test could be a reasonable alternative to the 24-hour urine collection for detection of significant proteinuria in hospitalized pregnant women with suspected preeclampsia. This test can really solve the problem of diagnosis of proteinuria in mild preeclamptic patients. Because of its advantage over dipsticks it can even routinely use to screen proteinuria in pregnant women.

The random urinary protein-to-creatinine ratio is a rapid, reliable and cost effective test, so this test was suggested for the purpose of screening, assessment or follow-up of proteinuria in the preeclampsia. So we recommend that future study should perform in outpatient basis and to evaluate clinical outcome when using this test for follow-up of proteinuria in preeclamptic patients.

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