**Original Research Article**

# A comparison of spot urinary protein-creatinine ratio with 24-hour urine protein in quantification of proteinuria in women with hypertensive disorders of pregnancy

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## ABSTRACT

**Background:** Proteinuria is a major indicator of hypertensive disorder of pregnancy. The gold standard for diagnosis of significant proteinuria is based on a 24hour urine collection which is cumbersome, time-consuming, and inconvenient to patients. A need, therefore, exists for a rapid, valid, and accurate test to identify significant urinary proteinuria.
**Methods:** We conducted an observational cross-sectional hospital-based study wherein a total of 150 pregnant women who met the criteria of preeclampsia were taken. Spot urine samples for calculating the urine P/C ratio were taken immediately before 24hr urine collection. Correlation between 24hour urine protein and spot PCR was then determined by Pearson's correlation coefficient(r).
**Results:** The correlation coefficient (r) between 24hr urine protein and spot PCR was found to be 0.734 (n=150) which was highly significant with p<0.001. In this study, population the ROC curve analysis revealed the sensitivity of 90.3% and specificity of 97.3% with AUC 0.958 for a cut-off value of spot PCR as 0.43 to detect significant.
**Conclusions:** Spot PCR is an accurate, valid, steady fast, reliable, and time-saving test which may be used as an alternative method for determining significant proteinuria in patients with pregnancy-induced hypertension.

**Keywords:** Preeclampsia, Proteinuria, Spot PCR, 24hours urine protein

## INTRODUCTION

Pregnancy-induced hypertension is the most frequent medical complication of pregnancy and is a major cause of perinatal mortality and morbidity. Among the hypertensive disorders of pregnancy, preeclampsia remains the leading cause that complicates 2-8% of all pregnancies.[1,2] Pregnancies complicated with hypertension result in a high incidence of hospital admissions, labor inductions, and fetomaternal morbidity and mortality. Proteinuria is a major indicator of hypertensive disorder of pregnancy, progresses with the course of disease and its presence indicates worsening of the hypertensive disorder.

Hence, it is essential to diagnose proteinuria accurately. Significant proteinuria is defined by the international society for the study of hypertension in pregnancy as excretion of ≥300 mg of protein in a 24-h urine specimen. Thus, the gold standard for diagnosis of significant proteinuria is based on a 24-h urine collection which is cumbersome, time-consuming, and also inconvenient to patients as well as hospital staff. Often incomplete collection leads to inaccuracies (13-68%).[4] A need therefore, exists for a rapid, accurate as well as valid test

to identify significant urinary proteinuria. This may lead to decision making thereby reducing patients anxiety, shorten hospital stay with its associated cost savings, and targeting those women with true pathology for treatment.

The usefulness of this study is much more relevant because there is dearth of information in literature regarding the accuracy of spot UPCR and its recommended cut-off in the Indian population. The clinical relevance of urinary spot protein-creatinine ratio as a substitution of 24hour urinary protein (gold standard) excretion for detecting significant proteinuria in patients with hypertensive disorders of pregnancy remains unclear. Keeping this in mind the present study was planned to assess whether measurement of urine protein:creatinine ratio in a single urine specimen in clinical practice may provide a reliable estimate of significant proteinuria (≥300 mg/24 h) in women with pregnancy-induced hypertension in clinical practice and also to find diagnostic accuracy of spot PCR.

## METHODS

This was an observational cross-sectional study. This study was carried out in pregnant women with hypertension attending the Department of Obstetrics and Gynaecology, Jawaharlal Nehru Medical College, AMU, Aligarh. Eligible Patients were enrolled from November 2019 to October 2022. The study was approved by the Institutional Ethical committee. Total 150 eligible patients were recruited during the study.

### Inclusion criteria

Inclusion criteria were the singleton pregnancy, age range between 20 and 40 years, gestational age ≥20 weeks, all patients with BP ≥140/90mmHg on at least two occasions taken 4hours apart, proteinuria of ≥1+ as detected by dipstick test.

### Exclusion criteria

Exclusion criteria were history of chronic hypertension, development of proteinuria before conception, chronic renal disease, pathological vaginal discharge, bleeding per vaginum, history of recurrent urinary tract infections, patients who require delivery before completion of collection of 24hr urine sample, multiple pregnancies.

A thorough general and obstetric history were taken for every enrolled patient. Informed consent was obtained from all enrolled patients. Every patient was assessed with respect to general physical, systemic, and obstetric examination. All routine antenatal investigations (blood group typing, hemoglobin estimation, liver function test, kidney function test, urine routine examination, HIV testing, HBsAg testing, STS) were performed on admitted patients. Spot urinary protein creatinine ratio was conducted in all patients. Additionally, 24-hour urine protein estimation was carried out in all patients as a gold standard test. Collection of 24-hour urine was begun in the morning after discarding the first voided urine of morning. Patients were directed to collect a second midstream urinary sample for evaluating spot urinary protein:creatinine ratio. The sample was collected in a clean bottle and patient was instructed to clean the perineal area with saline before sample collection.

The samples were analysed for the following: 1) Measurement of 24hour urine protein was done by Esbach's method wherein an aliquot is taken from 24hour urinary sample and the reagent (10gm picric acid and 20gm citric acid to 1L distilled water) is added to it.[5,6] Compound is left standing for 24hours then. Precipitated protein is interpreted in gm/litre, 2) Urine creatinine was detected by modified Jaffe's kinetic method, 3) The urine protein and creatinine ratio was obtained by dividing the urine protein concentration (mg/dl) by the urine creatinine concentration (mg/dl).

### Statistical analysis

Collected data were entered into Microsoft excel and then into SPSS. Demographic data used descriptive statistics and was presented as mean±SD, median and percentage, wherever appropriate. The relationship between the urine protein creatinine ratio and 24-hour protein excretion was estimated with Pearson's correlation test and the correlation coefficient was calculated which was expressed as "r". Inferential statistics were applied using chi-square test. Sensitivity and specificity were determined and the cut off value for spot PCR was calculated by the ROC curve.

## RESULTS

The mean period of gestation was 34.76±3.5weeks (25-40weeks) and mean age of patients under study was 24.97±4.0years (20-37years). The mean systolic blood pressure at the time of recruitment was 145.5±6.9mmHg and the mean diastolic blood pressure at the time of recruitment was 93.81±5.3mmHg, after 4 hours mean SBP was 145.44±5.4mmHg and mean DBP was 93.3±4.8mmHg, respectively. The incidence of preeclampsia in primigravida was 60% and mutigravida was 40% in this study (Table 1).

**Table 1: Demographic characteristics of study population.**

| Mean age | 24.97±4.0years |
|---|---|
| Mean parity | Primigravida-60% |
| | Multigravida-40% |
| Mean gestational age | 34.76±3.5 weeks |
| Mean systolic pressure during recruitment | 145.5±6.9mmHg |
| Mean diastolic pressure during recruitment | 93.81±4.8 |
| Mean systolic pressure after 4h | 145.44±5.4 |
| Mean diastolic after 4h | 93.3±4.8 |

The mean spot PCR was 0.99±0.98 and the mean 24hour urinary protein was 675.2±528.6. The correlation coefficient (r) between 24hour urine protein and spot PCR was found to be 0.734 (n=150) which was highly significant with p<0.00 (Table 2).

**Table 2: Correlation between 24-hour urinary protein and spot PCR.**

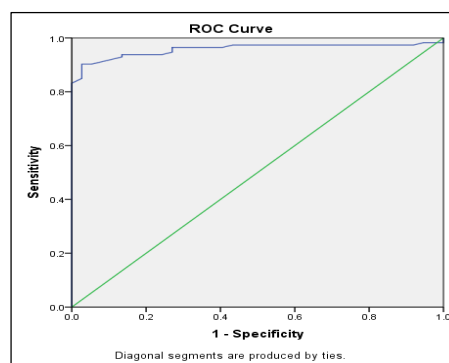| Correlations | | 24 HUP | Spot PCR |
|---|---|---|---|
| **24 HUP** | Pearson Correlation (r) | 1 | 0.734** |
| | P-value | | .000 |
| | N | 150 | 150 |
| **Spot PCR** | Pearson Correlation (r) | 0.734** | 1 |
| | P- value | .000 | |
| | N | 150 | 150 |

Receiver operator curve was plotted for spot urine PCR depicting its sensitivity and specificity for predicting significant proteinuria. Area under ROC curve is 0.958 for UPCR cut value of 0.43 (Figure 1) to predict 24hour urine protein of ≥300mg/24hour and 0.94 at UPCR cut off of 1.83 (Figure 2) to predict proteinuria of 2000mg/24hour respectively. The ROC curve analysis revealed a sensitivity of 90.3% and specificity of 97.3% with AUC 0.95 for a cut off value of spot PCR as 0.43 to detect significant proteinuria of >300mg/24hour and similarly to detect proteinuria of >2000mg/24hour, the cut off value of spot PCR is 1.83 with sensitivity of 100% and specificity 91.2% (Table 3).



**Figure 1: ROC curve for UPCR to predict 300mg/24hour proteinuria.**
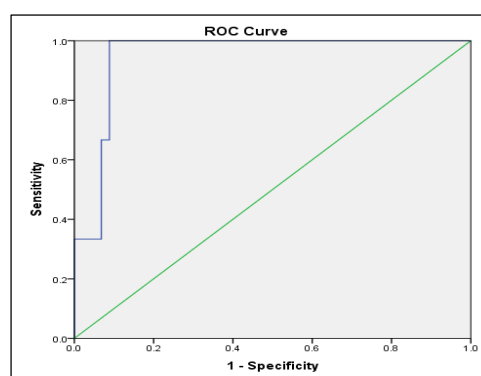


**Figure 2: ROC curve for UPCR to predict 2000mg/24hour proteinuria.**

**Table 3: Diagnostic ability of urine protein: creatinine ratio (UPCR) for various proteinuria range.**

| Proteinuria range | Cut off values | Sensitivity (%) | Specificity (%) | Accuracy (%) | Area under curve and 95% CI |
|---|---|---|---|---|---|
| **UPCR to predict 300 mg+/day** | 0.43 | 90.3 | 97.3 | 95.8 | 0.958 (0.926-0.990) |
| **UPCR to predict 2000 mg+/day** | 1.83 | 100 | 91.2 | 94.8 | 0.948 (0.89-1.00) |

**DISCUSSION**

An accurate and rapid quantitation of urinary protein is important in the management of hypertensive disorders in pregnancy. This can help us know the severity of proteinuria and the disease process, which can alter the course of management. Several investigators have explored other means of quantifying proteinuria in a shorter period. In this study, we enrolled 150 pregnant women with pregnancy-induced hypertension attending antenatal clinics, admitted to ward and labor room in the Department of Obstetrics and Gynecology. This study was limited to hospitalized non-ambulatory patients. Postural changes influence the excretion of proteins as the excretion is more in standing than in supine position and so the ambulatory status of the patient is important while

estimating results. Other means of quantifying proteinuria in a shorter period have been reported by several studies. In our observational cross-sectional study, we aimed to find diagnostic accuracy of spot urinary protein:creatinine ratio in the quantification of proteinuria in women with hypertensive disorders of pregnancy.

In our study, we found the mean age of studied patients as 24.97±4.0 years and the mean gestational age as 34.76±3.5 weeks. Similarly in the study done by Aggarwal et al, the mean age of patients was 26.30 years, and the mean gestational age of studied patients was 36weeks.[7] Amin et al also reported mean age and gestational age as 27.4±4.3 years and 35.3±3.3 weeks respectively.[8] These findings were in concordance with the study done by Stefanska et al who observed mean age was 28±4.05 years

for preeclampsia patients and the mean gestational age was 35 weeks.[9]

Neithardt et al who evaluated 30 pregnant patients with pregnancy- induced hypertension reported the average age of patients as 29.4 years.[10]

In this study, we found a correlation coefficient of 0.734 between 24hour urinary protein and spot PCR, which was highly significant at $p<0.001$(n=200). The ROC curve analysis revealed a sensitivity of 90.3% and specificity of 97.3% with AUC 0.95 for a cut off value of spot PCR as 0.43 to detect significant proteinuria of >300mg/24hour and similarly to detect proteinuria of >2000mg/24hour, the cut off value of spot PCR is 1.83 with sensitivity of 100% and specificity 91.2%.

Study results were in agreement with the study done by Nischintha et al in 75 patients which showed a moderate correlation between 24-hour urine protein and spot urine P/C ratio ($r = 0.38$) with a $P = 0.001$.[11] The ROC curve analysis in their study revealed a sensitivity of 73.53% and specificity of 65.85% with AUC 0.80 (good test) for a cut-off value of spot P/C>0.60 to detect significant proteinuria.

Aggarwal et al in 2004 reported a significant association between the two tests with a correlation coefficient of $r = 0.60$ ($P<0.01$) that is, a moderate correlation and the sensitivity and specificity of spot urine PCR at a cut-off value >1.14 of 72% and 75% respectively.[12]

There have been conflicting reports too. Durnwald and Mercer, in their study, reported a poor correlation with a coefficient of 0.41 between 24-hour urine and spot urine PCR.[13] The ROC analysis revealed no clear shoulder although the AUC was 0.80 with a sensitivity of 55.8% and specificity of 81% at a cut-off value of 0.30 for spot urine PCR. Furthermore, Durnwald and Mercer also recruited outpatient participants, who could have incomplete urine collections.

Because of the variability in laboratory methods for measuring proteinuria in different reported studies, several cutoff points and different units for the urinary PCR have been reported, thereby precluding valid comparisons among such studies. The high specificity shown by spot urine PCR in our population signifies that it will accurately diagnose preeclampsia and prevent unnecessary interventions.

Current study was also in concordance with the study by Taherian et al done in 2006, in which it was found that the cutoff value of >0.18 yields a sensitivity of 86.3% and a specificity of 100%.[14] In another study by Al et al, done to study the use of random urine protein-creatinine ratio for predicting proteinuria in new-onset mild hypertension in late pregnancy in 185 women, the recommended cutoff for spot urine PCR to detect significant proteinuria was 0.19 with the sensitivity of 85% and specificity of 73%.[10]

Differences in cutoff values could be due to variations in urine sample collections, participant demographics, sample size, inclusion, and exclusion criteria, and other effective factors. Since the protein excretion is influenced by postural change, being higher in the standing than in the supine position, the ambulatory status of the patients was also given importance while interpreting the results. Furthermore, spot urine protein creatinine ratio estimation can be performed on a similar biochemical analyzer as the one used for 24-hour urine protein estimation which was the same analyzer used for running routine urgent biochemical tests. Hence, it was found to be very cost-effective and feasible. Also, repeating a random sample is much easier and quicker to accomplish. By employing appropriate and strict exclusion criteria, we eliminated false high urine protein values. However, as our data was slightly skewed and did not necessarily represent all age groups equally, there may be some limitations to it.

Therefore, we conclude that the cutoff for spot urine protein creatinine ratio for detecting significant proteinuria in our population could be 0.43. Although spot urine protein creatinine ratio is an excellent test to rule out preeclampsia, the women with abnormal spot urine PCR should undergo 24-hour urine protein before clinical intervention is done. Further studies in patients with severe proteinuria are needed.

## CONCLUSION

In this study, there was a strong correlation between spot urine PCR and 24-hour urine protein with r equal to 0.734, $p <0.001$ (n=150). We also found that the cut-off for spot urine PCR in this study population would be 0.43 for proteinuria >300mg/24 hour and 1.83 for proteinuria of 2000mg/24hour. Thus, spot PCR may be used as an alternative to 24hour urine protein in women with pregnancy-induced hypertension in clinical practice.

## REFERENCES

1. MacKay AP, Berg CJ, Atrash HK. Pregnancy-related mortality from preeclampsia and eclampsia. Obstet Gynecol. 2001;97(4):533-8.
2. Rochat RW, Koonin LM, Atrash HK, Jewett JF. Maternal mortality in the United States: report from the Maternal Mortality Collaborative. Obstet Gynecol. 1988;72(1):91-7.
3. Shah S, Sulthana P. A comparative study of urine dipstick and urine protein creatinine ratio with 24 hour urinary protein in estimation of significant proteinuria in pre-eclampsia. IOSR. 2019;18(7):21-9.
4. Côté AM, Firoz T, Mattman A, Lam EM, von Dadelszen P, Magee LA. The 24-hour urine

collection: gold standard or historical practice?. Ame J Obstet Gynecol. 2008;199(6):625-e1.

5.  Adhikari R, Sharma GD, Dhungana PR, Adhikari KG. Is urine dipstick as accurate as 24 hour urine protein? A comparative study. Med J Pokhara Acad Heal Sci. 2018;1(2):110-5.

6.  Mahesh S, Borgohain D. A study to find the correlation between 24-hour urinary protein and spot urinary albumin to creatinine ratio and to determine the accuracy of spot urinary albumin to creatinine ratio as an indicator to detect proteinuria and its use as a rapid alternative test over 24-hour urinary protein in preeclamptic women. Inter J Reprod Contracep Obstet Gynecol. 2019;8(11):4358-66.

7.  Aggarwal A, Rathore A. Evaluation of spot urinary protein creatinine ratio for quantification of proteinuria in women with hypertension in pregnancy: A prospective observational study. Indian J Obstet Gynecol Res. 2020;7(2):163-7.

8.  Amin SV, Illipilla S, Hebbar S, Rai L, Kumar P, Pai MV. Quantifying proteinuria in hypertensive disorders of pregnancy. Inter J Hypert. 2014;2014:941408.

9.  Stefańska K, Zieliński M, Zamkowska D, Adamski P, Jassem-Bobowicz J, Piekarska K, et al. Comparisons of dipstick test, urine protein-to-creatine ratio, and total protein measurement for the diagnosis of preeclampsia. Inter J Environ Res Pub Heal. 2020;17(12):4195.

10. Neithardt AB, Dooley SL, Borensztajn J. Prediction of 24-hour protein excretion in pregnancy with a single voided urine protein-to-creatinine ratio. Ame J Obstet Gynecol. 2002;186(5):883-6.

11. Nischintha S, Pallavee P, Ghose S. Correlation between 24-h urine protein, spot urine protein/creatinine ratio, and serum uric acid and their association with fetomaternal outcomes in preeclamptic women. J Nat Sci Biol Med. 2014;5(2):255.

12. Agarwal I, Kirubakaran C. Quantitation of proteinuria by spot urine sampling. Ind J Clin Biochem. 2004;19(2):45-7.

13. Durnwald C, Mercer B. A prospective comparison of total protein/creatinine ratio versus 24-hour urine protein in women with suspected preeclampsia. Ame J Obstet Gynecol. 2003;189(3):848-52.

14. Taherian AA, Dehbashi S, Baghban HM. The relationship between random urinary protein-to-creatinine ratio and 24-hours urine protein in diagnosis of proteinuria in mild preeclampsia. JRMS. 2006;11(1):6-12.