

To evaluate the diagnostic value of protein: creatinine ratio in a single voided urine sample for quantitation of proteinuria compared to those of a 24-hour urine sample in patients with preeclampsia

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ABSTRACT

Objective

This study was done to evaluate the diagnostic value of protein: creatinine ratio in a single voided urine sample for quantitation of proteinuria compared to those of a 24 hour urine sample in patients with preeclampsia.

Methods

A prospective simple random sample study was done on the hypertensive pregnant women attending the antenatal clinic or admitted in Department of Obstetrics and Gynaecology. It included all women being evaluated for preeclampsia, regardless of the alerting sign or symptom, suspected severity or co-morbid conditions. The main measures were the urinary protein to urinary creatinine ratio by random (spot) direct measurement and the 24-h urinary protein excretion by a 24-h urine collection. The data obtained was statistically analyzed.

Results

Out of the 78 patients with gestational hypertension included in our study 48 patients had significant proteinuria ($e > 300$ mg/day). Only 2 patients had proteinuria of the range of greater than 3500mg. Among the patients, 50 had a positive protein: creatinine ratio ($e > 0.3$) while 28 patients had a negative protein: creatinine ratio (< 0.3). The P: C ratio was able to correctly identify 44 out of 48 patients with significant proteinuria (when the comparison is made with the gold-standard test; i.e., 24-hour urine protein). It could also identify 24 out of 30 patients without significant proteinuria as compared to the gold-standard test.

In this study, the Protein: Creatinine ratio with a sensitivity of 91.67%, a specificity of 80%, positive predictive values 88% and the negative predictive values 85.71%.

Conclusions

Our data suggests that the protein: creatinine ratio in single voided urine is a highly accurate test (p value < 0.0000001) for discriminating between insignificant and significant proteinuria. Based on the above findings we conclude that a random urine protein excretion predicts the amount of 24- hour urine protein excretion with high accuracy. This could be a reasonable alternative to the 24-hour urine collection for detection of significant proteinuria in hospitalised pregnant women with suspected preeclampsia.

Key words: preeclampsia, protein: creatinine ratio, single voided and 24 hour urine.

INTRODUCTION

Providers of obstetric care must be familiar with the diagnosis of pre-eclampsia, the hypertensive condition associated with the highest risk of adverse maternal and perinatal complications and a condition that affects 2-5% of pregnancies.^{1,2} Pre-eclampsia is defined as systolic blood pressure level of 140mm of Hg or higher or a diastolic blood pressure level of 90mm of Hg or higher that occurs after 20 weeks of gestation with proteinuria.³ Proteinuria is an important sign of preeclampsia, and diagnosis is questionable in its absence. Preeclampsia is an important cause of maternal morbidity and mortality as well as a significant contributor to increased perinatal morbidity and mortality rates in India. Twelve percent of all maternal deaths worldwide are due to hypertensive disorders of pregnancy.⁴

Urine collection over 24 hours is considered the traditional comparator for quantification of proteinuria in pregnancy, when significant proteinuria is defined as proteinuria of 300 mg/day or more.^{1,2,5,6} Patients with hypertension have only <300mg, those with mild pre-eclampsia have 300mg to 5000mg and those with severe pre-eclampsia have >5000mg of proteinuria. The urine requires refrigeration and its collection is cumbersome, time consuming (for women and ward staff), and potentially misleading if collected inaccurately. Also, it may not be possible to complete the urine collection when delivery occurs, leading to undetermined proteinuria status and an unsubstantiated diagnosis of pre-eclampsia; less than half of women admitted with pre-eclampsia have a 24 hour urine collection sent for analysis.^{1,7} Timed collections delay clinical diagnosis and may

result in prolonged hospital stay when a hypertensive disorder of pregnancy is being investigated, thereby increasing patient anxiety and healthcare costs.

Because of the disadvantages of 24 hour urine collection, alternatives for the diagnosis of proteinuria in pregnancy have been considered. These include urinary dipsticks, urine collections over a shorter period, the urinary spot protein: creatinine ratio, and the urinary spot albumin: creatinine ratio. 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.^{8,9,10}

The spot protein: creatinine ratio and spot albumin: creatinine ratios have been well studied and used outside pregnancy.

Recently, the urine protein–creatinine ratio has been considered important for predicting proteinuria in pregnant patients. It compares the spot urine protein excretion to the spot urine creatinine excretion, thereby normalizing protein excretion to the glomerular filtration rate. Thus, the urine protein–creatinine ratio is not subject to variation due to hydration status. In pregnant women, the urine protein–creatinine ratio and the 24-h urine are highly correlated.^{11,12,13} We carried out a systematic review to assess the diagnostic value of protein: creatinine ratio in a single voided urine samples compared with 24 hour urinary collection for the detection of significant proteinuria in hypertensive pregnant women attending the antenatal clinic or admitted in the antenatal ward of Calcutta National Medical College and Hospital.

OBJECTIVE

To evaluate the diagnostic value of protein: creatinine ratio in a single voided urine sample for quantitation of proteinuria compared to those of a 24 hour urine sample in patients with preeclampsia.

METHODS

STUDY AREA AND PERIOD:

The study was performed in the Department of Obstetrics & Gynaecology, Calcutta National Medical College & Hospital from August 2010 to July 2011.

STUDY POPULATION:

Study population consisted of Booked/ Un-booked ambulant antenatal cases of gestational age >20 weeks with new onset Hypertension (defined as blood pressure \geq 140/90 mm of Hg in two different measurements obtained at interval more than 6 hours).

Subjects were excluded if they had concurrent diseases like: Chronic hypertension, Diabetes mellitus, Bacteriuria, Urinary tract infection, Renal disorder. Women receiving diuretics, who had history of vulval cleansing with antiseptics or chlorhexidine and if the urinalysis contained >10 WBCs per h.p.f. or if a catheter was not used after membrane rupture were also excluded. Complications of preeclampsia, patients who delivered before completion of collection of 24-hour urine sample, who were on bed rest longer than 24 hours or performed heavy exercise (> 1hour of vigorous exercise on the day of urine collection) were also excluded.

SAMPLE SIZE

78 women with gestational hypertension.

STUDY DESIGN

Prospective study with Simple random sample.

PARAMETERS STUDIED:

24-hour urine protein and Protein: creatinine ratio in single voided urine sample.

STUDY TECHNIQUE:

The study was done on the hypertensive pregnant women attending the antenatal clinic or admitted in the antenatal ward of Calcutta National Medical College and Hospital. It included all women being evaluated for preeclampsia, regardless of the alerting sign or symptom, suspected severity or co-morbid conditions. The main measures were the urinary protein to urinary creatinine ratio by random (spot) direct measurement and the 24-h urinary protein excretion by a 24-h urine collection. 24-hour urine collection was started before midday. First morning sample was discarded and the time was noted. Urine was collected in a clean bottle for 24 hours and the last sample was taken on the next day at the same time. A single voided urine specimen for spot protein: creatinine ratio was obtained as soon as possible after the 24-h collection. All samples were collected via clean catch unless the membranes had been ruptured, in which case specimens were obtained by catheter. Morning sample was excluded for estimation of urine P: C ratio. A complete collection was defined as having a total creatinine of >1000 mg (850 mg for obese women) or a total creatinine of 13 mg per kg body weight.²⁰

RESULTS

TABLE 1: Distribution of the study subjects (n=78) according to Age

AGE (IN YEARS)	NUMBER (n)	PERCENTAGE (%)
<20	17	21.8
20-29	55	70.5
e"30	6	7.7
TOTAL	78	100

Most of the patients in our study population were in the 20-29 years age group (70.5%). 21.8% were in the teenage group and only 7.7% patients belonged to e"30 years group in our study.

TABLE 2: Distribution of the study subjects (n=78) according to Parity

ARITY	NUMBER (n)	PERCENTAGE (%)P
0	61	78.2
1	14	18
2	2	2.5
e"3	1	1.3
TOTAL	78	100

Our study revealed that 78.2% patients were nulliparous where as the rest were multiparous. Out of the 17 multipara, 14 (18%) were primirara, 2 were para 2, and only one a para 3.

TABLE 3: Distribution of the study subjects (n=78) according to 24-hour urine protein

24 HOUR URINE PROTEIN(gm/day)	NUMBER (n)	PERCENTAGE (%)
<0.3	30	38.5
0.3-3.5	46	59
>3.5	2	2.5
TOTAL	78	100

Out of the 78 patients included in our study 48 patients had significant proteinuria (e"0.3gm/day). Only 2 patients had proteinuria of the range of greater than 3.5g/day.

TABLE 4: Distribution of the study subjects (n=78) according to Protein: Creatinine ratio in spot urine sample

PROTEIN: CREATININE RATIO in spot urine sample	NUMBER (n)	PERCENTAGE (%)
<0.3	28	35.9
0.3-0.35	8	10.2
>0.35	42	53.9
TOTAL	78	100

Among the 78 patients in the study population, 28 (35.9%) had a protein: creatinine ratio less than 0.3, 8 (10.2%) had a ratio between 0.3 and 0.35 while 42 (53.9%) patients had a ratio of greater than 0.35.

TABLE 5: Comparison of Protein: Creatinine ratio in single voided urine sample to 24-hour urine protein in the study subjects (n=78)

P:C RATIO IN SINGLE VOIDED URINE SAMPLE	24 HOUR URINE PROTEIN		TOTAL(n)
	POSITIVE (e"300mg)	NEGATIVE (<300mg)	
POSITIVE P:C(e"0.3)	44(TP)	6(FP)	50
NEGATIVE P:C(<0.3)	4(FN)	24(TN)	28
TOTAL	48	30	78

TP= True Positive
FP= False Positive
TN= True Negative
FN= False Negative

Out of the total 78 patients with gestational hypertension, 48 patients had a 24-hour urine protein excretion greater than or equal to 300mg. Out of these 48 patients the protein: creatinine ratio was positive (i.e., e"0.3) in 44 patients; which signifies that these 44 patients were the true positive subjects in the study while the 4 study subjects who had a negative P: C ratio (<0.3) were the false negative subjects. In our study, 30 patients with gestational hypertension had 24-hour urine protein excretion less than 300mg. Out of them the P: C

ratio was negative (<0.3) in 24 patients who were the true negative study subjects; and 6 had a positive P: C ratio (e"0.3) that is they were the false positive study subjects.

Thus, we can say that the P: C ratio is able to correctly identify 44 out of 48 patients with significant proteinuria (when the comparison is made with the gold-standard test; i.e., 24-hour urine protein). It can also identify 24 out of 30 patients without significant proteinuria as compared to the gold-standard test.

TABLE6: Diagnostic value of Protein: Creatinine ratio in patients with suspected preeclampsia (n=78) compared to 24-hour urine protein

TEST OF VALIDITY	PROTEIN : CREATININE RATIO
SENSITIVITY	91.67%
SPECIFICITY	80%
POSITIVE PREDICTIVE VALUE	88%
NEGATIVE PREDICTIVE VALUE	85.71%
FALSE POSITIVE RATE	20%
FALSE NEGATIVE RATE	8.33%

The sensitivity, specificity, positive predictive value, negative predictive value, false positive rate and false negative rate of the study were calculated from results of table no 5.

The Protein: Creatinine ratio in our study had a sensitivity of 91.67% which allows the clinician to correctly identify greater than 9 out of 10 cases of significant proteinuria. The Protein: Creatinine ratio had a specificity of 80% in our study population. The positive predictive value of the urine Protein: Creatinine ratio was 88%. The negative predictive value of the test was 85.71%. The study had a false positive rate of 20% and a false negative rate of 8.33%.

Data analysis was done by the EPI Info 3.2 Software which revealed:

Odds Ratio 44 (9.74 < OR < 226.75)

Confidence Limit of 95%.

Chi – Square Test:

$\chi^2 = 38.15$

Yate's corrected

$p = 0.00000001$, which is highly significant.

DISCUSSION

Most of the patients of our study population were in the 20-29 years age group (70.5%). 21.8% were in the teenage group and only 7.7% patients belonged to age group of greater than or equal to 30 years. The peak age of 20-29 years may be reflective of the fact that most first deliveries in the Indian scenario occur at that age and not necessarily of any special contribution of this age bracket to the aetiology of the disease.

Various studies have shown that women aged e" 40 had approaching twice the risk of developing pre-eclampsia,¹⁴ whether they were primiparous or

multiparous (relative risk 1.68, 95% confidence interval 1.23 to 2.29, and 1.96, 1.34 to 2.87, respectively). Nationwide US data suggest that the risk of pre-eclampsia increases by 30% for every additional year of age after 34 years.¹⁵ Young maternal ages did not seem to affect the risk of developing pre-eclampsia, whichever cut off age was used.

Out of the 78 patients included in our study 48 patients had significant proteinuria. Only 2 patients had proteinuria of the range of greater than 3500mg. Among the patients, 28 had a protein: creatinine ratio less than 0.3, 8 had a P: C ratio between 0.3 and 0.35 while 42 patients had a P: C ratio of greater than 0.35.

Several investigators have explored other means of quantifying proteinuria in a shorter period. In this study a comparison of the Protein: Creatinine ratio with the standard 24 hour protein estimation using the various indices of validity was quite revealing. Sensitivity, Specificity, Positive and Negative predictive values, true and false positive rates were the indices used. The odds ratio and p value were also calculated.

In this study, the Protein: Creatinine ratio with a sensitivity of 91.67% allows the clinician to correctly identify greater than 9 out of 10 cases of significant proteinuria.

The Protein: Creatinine ratio had a specificity of 80% in our study population. The high specificity shown by the Protein: Creatinine ratio will accurately diagnose preeclampsia and prevent unnecessary interventions.

The study had a false positive rate of 20%. Numerous studies have demonstrated that false

positive reactions may occur with concentrated urine, highly alkaline urine (pH > 8), contamination of urine with vaginal discharge and antiseptics like chlorhexidine. The false positive rate for the spot urine Protein: Creatinine ratio of 20% still leaves some room for errors in diagnosis and premature intervention.

It has been suggested that sensitivity and specificity are not as useful to the clinician as the positive and negative predictive values of the tests. This is because the sensitivity and specificity (population measures) look backward at results gathered over time; clinicians have to interpret test results to those tested. Thus, what clinicians need to know are the predictive values of the tests.

The clinician and the patient need to know what the probability is that a positive result is genuinely positive (Positive Predictive Value) and what probability that a negative result is genuinely negative (Negative Predictive Value). This determines the confidence the clinician has in a positive or negative result and his willingness to base clinical judgements on the results. The study reveals that the positive predictive value of the urine Protein: Creatinine ratio to be 88% and the negative predictive value to be 85.71%.

Our data suggested the protein: creatinine ratio in single voided urine is a highly accurate test (p value < 0.0000001) for discriminating between insignificant and significant proteinuria, independent of gestational age, parity, height, weight and the degree of proteinuria.

Many studies have shown a high correlation between random urinary protein: creatinine ratio in both normotensive and hypertensive pregnant

women,¹⁶ and pregnant and non-pregnant patients with diabetes mellitus and underlying renal disease,¹⁷ This correlation is also showed in the present study. Correlation coefficients reported range from 0.80 to 0.995, and the degree of correlation did not vary by trimesters of pregnancy during which the sampling occurred¹⁸. Despite the high degree of linear correlation, the best cut off has not been described for pregnancy, and the test is not widely used during pregnancy.^{18,19}

Our study population was ambulatory. Also it avoided the potential impact of prolonged bed rest before sampling on the protein: creatinine ratio. Our study population was clinically appropriate. For patients with a high pretest probability of disease and a negative random urinary protein to creatinine ratio, repeating the test or proceeding with collection of a 24-hours urine is a reasonable option. Repeating a random sample is much easier and quicker to accomplish. In present study, the random urinary protein: creatinine ratios relationship (as an easier method for evaluation of proteinuria in women with suspected preeclampsia) with the 24-hours protein excretions have been evaluated and show an excellent correlation, independent of gestational age, parity, height, weight and the degree of proteinuria. Besides, it decreases hospital cost and patient inconvenience, especially when frequent or even daily determinations

CONCLUSIONS

In our study, the Protein: Creatinine ratio had a sensitivity of 91.67% which will allow the clinician to correctly identify greater than 9 out of 10 cases of significant proteinuria. The high specificity of 80% shown by the test will help to accurately

diagnose women with preeclampsia and prevent unnecessary interventions. Our data suggests that the protein: creatinine ratio in single voided urine is a highly accurate test (p value < 0.0000001) for discriminating between insignificant and significant proteinuria. Based on the above findings we conclude that a random urine protein excretion predicts the amount of 24- hour urine protein excretion with high accuracy. This could be a reasonable alternative to the 24-hour urine collection for detection of significant proteinuria in hospitalised pregnant women with suspected preeclampsia. The results of this study demonstrate that in hospital with appropriate laboratory personnel and where patients can afford it, routine use of the protein: creatinine ratio for quantitation of proteinuria in patients with preeclampsia could be adopted. The protein: creatinine ratio especially is reliable, relatively faster and accurate for proteinuria correlating with 24 hour urinary protein excretion.

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