COMPARISON OF ESTIMATED PROTEIN EXCRETION RATE WITH PROTEIN CREATININE RATIO IN RELATION TO 24 HOUR URINARY PROTEIN IN HEALTHY INDIVIDUALS

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ABSTRACT

Background: Protein-to-Creatinine Ratio (PCR) is used as an alternative to 24-hour urinary protein excretion. PCR has only one drawback that it cannot assess the nephrotic range proteinuria accurately. Estimated protein excretion rate can be an alternative to PCR in cases where PCR fails to detect high grade proteinuria.

Objective: To assess diagnostic ability of estimated protein excretion rate in relation to 24 hour urinary protein as compared to protein to creatinine ratio in healthy individuals.

Methods: It was an analytical cross sectional study conducted at Pathology department of Rehman Medical Institute Peshawar from August 2021 to July 2022. Seventy five healthy subjects from both genders who were potential donors of renal transplant were selected by non-probability consecutive sampling technique. Urine samples for spot PCR were taken after patients were done with their 24-hours urine collection. Estimated Protein Excretion Rate (ePER) was calculated by using PCR and estimated Creatinine Excretion Rate (eCER) equation. Twenty four hour urinary protein, PCR and ePER were expressed as medians and interquartile ranges (IQRs). Spearman correlation was applied to assess the correlation among 24-hour urinary protein loss, PCR and ePER.

Results: Mean age of the subjects was 33.75 ± 13.75 years. On categorization of subjects for proteinuria, according to PCR, out of 75, 70 subjects were sub-nephrotic and 5 were nephrotic while according to 24-hour protein excretion, only 30 subjects were sub-nephrotic, 1 nephrotic and 44 were normal. On spearman correlation, PCR, ePER (males) and ePER (females) showed significant correlation (<.001) with 24 hour urinary protein.

Conclusion: The study could not prove that ePER is superior to PCR in relation to twenty four hour urinary protein. Both ePER and PCR can equally predict the proteinuria in relation to twenty four hour urinary protein loss.

Keywords: Protein-to-Creatinine Ratio, estimated Protein Excretion Rate, Sub-nephrotic proteinuria, Nephrotic proteinuria.

INTRODUCTION

Proteinuria is an important indicator for the assessment of risk of renal disorders in general population as well as in patients having chronic kidney disease (CKD) [1].

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Therefore its precise identification and estimation are important for the diagnostic as well as management purposes of chronic kidney diseases. Although twenty four hour urinary protein excretion is a gold standard for estimation of urinary protein excretion, yet protein/creatinine ratio is a reasonable alternative [2].

The protein-to-creatinine ratio correlates well with twenty four hour protein excretion and the cut-off values of PCR are recommended in clinical settings [3]. Spot PCR poorly predicts 24-hour proteinuria in the range of 300-2000 mg [4]. PCR in a spot urine sample in patients undergoing a kidney transplant is a convenient and reliable method of estimating urinary protein excretion.

Basically protein-to-creatinine ratio is a measure of protein excretion rate (PER), therefore considering urine creatinine excretion rate (CER) would help in establishing a correlation between PCR and twenty four hour

urine proteinuria [6]. Similarly urine albumin excretion rate (AER) can be estimated from Albumin to Creatinine Ratio (ACR), with the help of creatinine excretion rate [7].

The PCR and ACR use urine creatinine for estimation, but, creatinine excretion is also affected by muscle mass, which means that PCR may become biased due to differences in muscle mass [8]. Studies have concluded that older age, lower body weight, and female gender are associated with a higher PCR and ACR due to lesser muscle mass in these individuals, leading to lower urine creatinine excretion rate[9].

Joseph and colleagues suggest that if one multiplies first morning void ACR by the creatinine excretion rate (eCER) for calculating the estimated albumin excretion rate (eAER), it will decrease the bias due to variability in creatinine excretion between individuals, leading to a more accurate estimation of AER[10].

Hong et al in a study conclude that estimated Protein Excretion Rate (ePER) is superior to just PCR regarding estimation of daily proteinuria [11]. The current study therefore aimed to evaluate the significance of ePER in predicting proteinuria in comparison to 24-hour urinary protein estimation.

METHODOLOGY

It was an analytical cross sectional study and conducted at Pathology department of Rehman Medical Institute Peshawar from August 2021 to July 2022. The study was approved by the institutional ethical review board. A total of 75 subjects were included in this study. Sample size was calculated according to a similar study conducted by Hong et al[11]. Subjects of both genders not having history of CKD or acute kidney disease (AKI) and urinary tract infection were included in the study. These individuals were basically potential healthy donors for the candidates of kidney transplant. After informed verbal consent, samples for spot urinary protein/ creatinine ratio were taken from patients, right after patients were done with their twenty four hour urine collection. Urinary protein estimation was done on Cobas 501 analyzer by the turbidimetric method while creatinine was estimated on the same analyzer by the kinetic colorimetric method according to Jaffe's principle. Daily protein excretion was measured from 24-hour urine samples. Estimated Protein Excretion Rate (ePER) was estimated by multiplying PCR with estimated Creatinine Excretion Rate (eCER) [7]. The $eCER_{MDRD}$ both for males and females were derived from MDRD equation [12]. On the basis of $eCER_{MDRD}$, $ePER_{MDRD}$ for both males and females were calculated.

$$ePER_{MDRD} = PCR \times eCER_{MDRD}$$

$$eCER_{MDRD} \text{ (mg/d, male)} = 1307.3 + (23.1 \times age) - (0.3 \times age^2)$$

$$eCER_{MDRD} \text{ (mg/d, female)} = 1051.3 + (5.3 \times age) - (0.1 \times age^2)$$

Subjects were divided into three categories, according to 24-hour urinary protein excretion and spot protein to creatinine ratio [13].

Normal: Patients having urinary protein less than 150 mg/24 hour or 15 mg/g.

Sub-nephrotic: Patients having urinary protein 150-3500 mg/24 hour or 15-350 mg/g

Nephrotic: Patients having urinary protein more than 3500 mg/24 hour or 350 mg/g.

Statistical analysis was done by SPSS 23. Mean values of demographics like age and gender distribution were calculated and compared with each other by using independent t test. Different categories of proteinuria (on the basis of 24-hour protein loss and PCR) were compared by Chi square test. Twenty four hour urinary protein, PCR, eCER and ePER were subjected to Kolmogorov Smirnoff and Shapiro Wilk test to assess the normality of data. Data distribution was not normal, therefore these parameters were expressed as median and interquartile ranges (IQRs). Spearman correlation was used to assess the relationship among these variables. For the graphical presentation of correlations between these different variables, data were log transformed.

RESULTS

Mean age of the subjects was 33.75 ± 13.75 years. Number of males (42) was greater than females (33), similarly the mean age of males was 35.76 ± 15.48 while mean age of females was 31.18 ± 10.87 with no significant difference.

Table 1: Gender wise distribution of age of study population

Gender	Number	Mean age	Std. Deviation	P value
Male	42	35.76	15.48	0.154*
Female	33	31.18	10.87	

*= Independent t test

Out of 75 subjects, 70 were sub-nephrotic and 5 were nephrotic according to PCR categorization while on the basis of 24-hour urinary protein, 44 were normal, 30 were sub-nephrotic and 1 subject was nephrotic and there was a statistically significant difference among the PCR and 24-hour urinary protein categories.

Table 2: Categorization of subjects according to protein excretion

PCR categories	Number	TFHP categories	Number	P value
Normal	0	Normal	44	<0.001 *
Sub nephrotic	70	Sub nephrotic	30	
Nephrotic	5	Nephrotic	1	

TFHP: Twenty four hour protein, PCR: Protein to Creatinine Ratio, *= Chi square test

Median and Interquartile ranges of different parameters show that median values of ePER were greater in males as compared to females.

Table 3: Median and Interquartile ranges of different variables

Variable	Median	IQR
TFHP	135	95
PCR	85.6	72.43
ePER (males)	145357.90	118879.75
ePER (females)	95157.50	82733.20

TFHP: Twenty four hour protein, IQR: Interquartile range, ePER: Estimated Protein Excretion Rate

All the parameters were significantly correlated to each other. The correlation coefficient for twenty four hour protein and PCR was 0.467 indicating a moderate positive relationship. Similarly, the correlation coefficients between TFHP and PER for males and females were 0.477 and 0.461, respectively, both indicating a moderate positive relationship. On other hand PCR and ePER in males and females showed a strong positive relationship.

Table 4: Spearman correlation among different variables

Variable	Correlation coefficient (r^2)	P value
TFHP and PCR	0.467	<0.001*
TFHP and ePER (males)	0.477	<0.001*
TFHP and ePER (females)	0.461	<0.001*
PCR and e PER (males)	0.996	<0.001*
PCR and ePER (females)	0.994	<0.001*

TFHP: Twenty four hour protein,*= Spearman correlation, ePER: Estimated Protein Excretion Rate

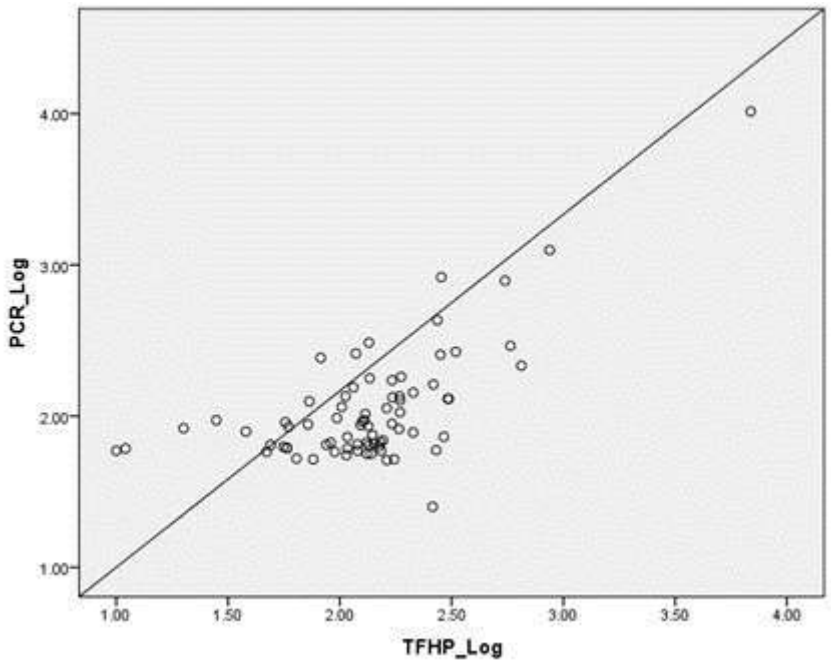

Figure 1: Correlation of log PCR to log 24-hour protein excretion

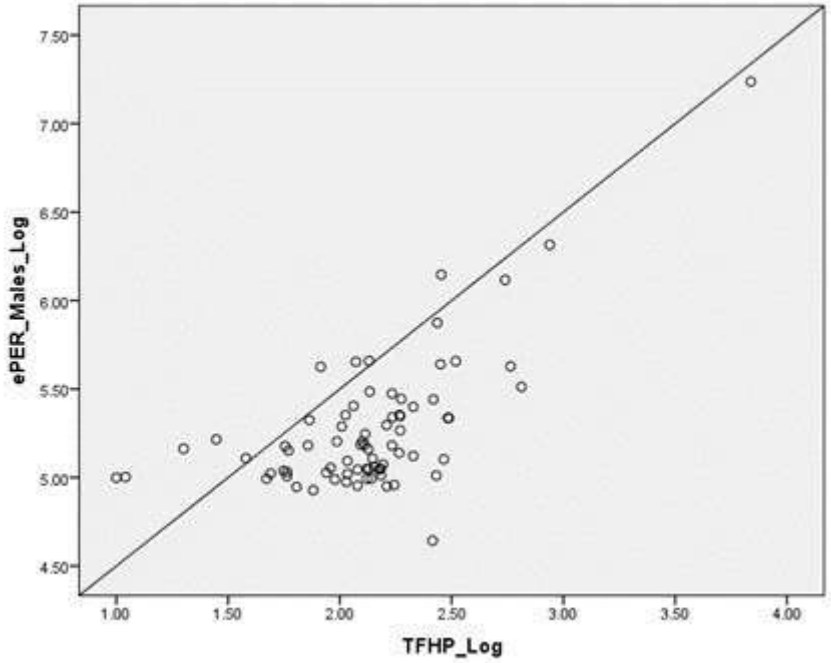

Figure 2: Correlation of log ePER (males) to log 24 hour protein excretion

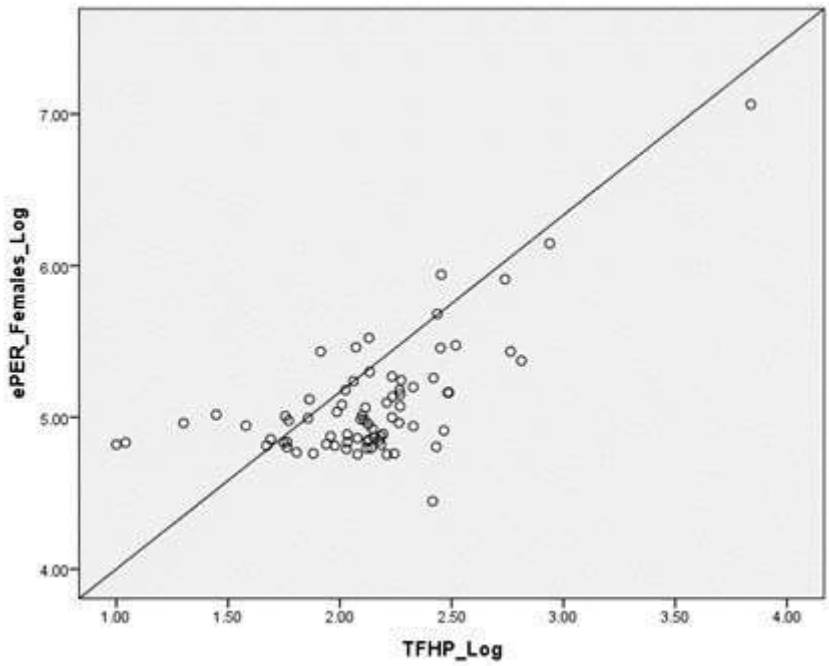

Figure 3: Correlation of log ePER (females) to log 24 hour protein excretion

DISCUSSION

In current study the number of males was slightly greater than females but without any statistically significant difference and this was in accordance to the result drawn by Hong et al [11].

On categorization of patients for proteinuria according to 24-hour urinary protein and Protein to Creatinine Ratio (PCR), it was found that PCR overestimated the degree of proteinuria. According to PCR, majority of subjects (70) were sub-nephrotic, 5 were nephrotic and no subject was in normal category. On the contrary according to 24-hour urinary protein, there were 44 subjects in normal category, 33 in sub-nephrotic and only one in nephrotic category. So, PCR overestimated the protein excretion. This finding was similar to the result derived by Sahu et al [14]. They concluded that out of 72 patients, 44 patients showed greater protein excretion on estimation by PCR when compared with protein excretion by 24-hour urinary protein.

The median values of ePER in males were greater than females. This finding is explainable by the fact that males have more muscle mass and therefore greater creatinine levels, greater estimated creatinine levels and ultimately grater ePER values.

On spearman correlation, PCR showed moderately significant correlation with 24-hour urinary protein, similarly ePER both in males as well as females also showed moderately significant correlation with 24-hour urinary protein. While PCR and ePER in males as well as females showed strongly significant correlation with each other. So PCR seemed equally significant as ePER for estimating 24-hour urinary protein loss in healthy individuals. These findings were in contrast to the results drawn by Hong et al [11] and Selvarajah and colleagues [15]. According to Hong et al ePER is superior to PCR for assessment of proteinuria while Selvarajah and colleagues concluded that Estimated Protein Output (EPO) may be marginally more accurate than PCR for the evaluation of proteinuria in first void urine sample.

The current study most probably shows different results from the studies in comparison, because those studies were conducted on subjects with chronic renal disease while the population of current study was healthy individuals. The reason, this study attempted to test the significance of PER over PCR in relation to 24 hour urinary protein was that PCR fails to predict the proteinuria accurately when it is in nephrotic range [16].

The reason for selecting healthy individuals instead of those with kidney disease was the fact that PCR in high grade proteinuria may overestimate protein excretion, therefore in order to reduce bias it was better to calculate ePER in healthy individuals.

CONCLUSION

There were positive correlations between estimated Protein Excretion Rate (ePER) and 24-hour urinary protein excretion. But these correlations were not more significant than that of PCR with 24-hour urinary protein. The present study could not find the ePER superior to PCR in relation to 24-hour urinary protein excretion in healthy individuals.

DECLARATIONS: Nil

Authors contributions:

1. Noman Shah: Conception of design, acquisition, and drafting and revising manuscript
2. Munir Husain: Literature search, acquisition, analysis and interpretation of data. Drafting and revision of manuscript
3. Bilal Iqbal: Drafting and revision of manuscript
4. Mohsin Shafi: Drafting and revision of manuscript
5. Mirza M Dawood: Manuscript revision, literature search and final approval of the version
6. Saman Hussain: Literature search, drafting and revision of manuscript

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Authors claim no conflict of interest

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