

The Correlation Between Protein Level in 24 Hour Urine Sample and Spot Urine Protein/Creatinine Ratio in the Elderly

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Abstract

Objective: This study aimed to determine whether there is a correlation between total protein level in 24-hour urine sample and protein-to-creatinine ratio in spot urine for measuring protein excretion in elderly patients and determining the reliable protein-to-creatinine ratio in spot urine threshold value for proteinuria.

Materials and Methods: Fifty in-patients, ≥ 65 years of age with a spot urine protein value ≥ 15 mg/dl measured by urine dipstick examination and had no risk factors for transient proteinuria were included in the study. Daily protein excretion was determined by two different methods: protein-to-creatinine ratio in spot urine and the total protein level in 24-hour urine sample. Correlation between these two methods were evaluated.

Results: There was a strong positive correlation between total protein level of 24-hour urine samples and protein-to-creatinine ratio in spot urine ($r=0.879$, $p<0.005$). When proteinuria level is ≥ 3.5 g/day, sensitivity and specificity of protein-to-creatinine ratio in spot urine also increased.

Conclusion: Protein-to-creatinine ratio in spot urine is a highly sensitive and specific test with high agreement with gold-standard method for proteinuria diagnosis and follow-up in elderly patients with chronic diseases. This will help clinicians for clinical decision making of elderly patients, especially when they are frail, have restricted mobility, incontinence or have difficulty in transfer secondary to medical, social or economical reasons.

Keywords: 24 hour urine, clinical geriatrics, protein/creatinine ratio, proteinuria, spot urine

Introduction

Proteinuria is an independent risk factor for cardiovascular and renal diseases and is an indicator of target organ damage. It is the most common clinical finding of underlying renal disease. In addition to being an early sign of renal disease, it is also a guide for differential diagnosis, determining prognosis and following treatment (1). The gold-standard method used to determine daily protein excretion in urine is the total protein level in 24-hour urine sample (24-HUP) (2, 3). The collection of 24-hour urine sample is a time-consuming method highly dependent on patient cooperation, frequently interfering with the health quality of the patient.

Besides, geriatric syndromes, including cognitive disorders, restricted mobility, incontinence and increased frequency of chronic diseases, both interfere with convenience, sensitivity and specificity of 24-HUP results and make sample collection more cumbersome for the elderly. Furthermore, it may also

increase hospital admission rates since the patient has to come at least once more to the clinics for leaving samples.

Protein-to-creatinine ratio in spot urine (PCR-SU) from an untimed specimen is the alternative method for proteinuria measurement (4). Since cheaper, more convenient and less time-consuming, PCR-SU is frequently used for the quantitative evaluation of proteinuria in practice (3, 5, 6).

Although there are studies that demonstrate a moderate-to-high correlation between 24-HUP and PCR-SU, this association is less evaluated in the elderly (7-9). This study aimed to determine whether there is a correlation between 24-HUP and PCR-SU for measuring protein excretion in elderly patients and determining the reliable PCR-SU threshold value for proteinuria.

Materials and Methods

All patients hospitalized between August 2015 and April 2016 in the Geriatrics Clinic of Ankara University Hospital were examined

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for inclusion into this study. All included patients were ≥ 65 years of age. Patients with urine pH > 8 , gross hematuria, presence of semen/leukocyte in the urine and history of iodinated contrast agent exposure in the last 24 hours were excluded since these may result in a false-positivity of proteinuria in the urine dipstick examination. Patients with urinary incontinence were also excluded since they would be unable to make a proper urine collection. Seventy-three in-patients were enrolled, of whom 23 were excluded [17 due to inadequate collection (missing some urine, pouring out, the wrong container), six due to over-collection]. Totally, 50 patients (27 males and 23 females) with proteinuria were included in the study. Daily urinary protein excretion was determined by two different methods: PCR-SU and 24-HUP. 24-hour urine samples were collected, excluding the first urine of the day and including the first-morning urine sample of the next day. Spot urine samples were taken as the first urine in the morning. The samples were collected during two consecutive days.

Total protein concentration levels were measured by a turbidometric assay using benzethonium chloride and creatinine level by Jaffe test in the Ankara University biochemistry laboratory. Serum creatinine levels were measured spectrophotometrically. The patients were categorized into three groups according to the Glomerular Filtration Rate (GFR) levels: GFR < 30 ml/min/1.73 m², GFR between 30-60 ml/min/1.73 m² and GFR > 60 ml/min/1.73 m² per the National Institute for Health and Care guidelines. The Modification of Diet in Renal Disease (MDRD) Formula was used for GFR calculations.

Excellence

All tests for this study were performed in the Ankara University biochemical laboratory, a standardized laboratory inspected regularly by the ministry of health of Turkey.

Statistics

All analyses were performed in Windows XP using SPSS version 22.0 (IBM Co., New York, USA). The Pearson correlation test was used to determine the relationship between the protein-to-creatinine ratio in spot urine and the protein levels in 24-hour urine sample. $P < 0.05$ was considered statistically significant. Differential thresholds, sensitivity and specificity of protein-to-creatinine ratio levels in spot urine were calculated using the receiver operator curves (ROC) curve for thresholds of ≥ 0.3 , ≥ 0.5 , ≥ 1 and ≥ 3.5 g/day proteinuria in 24-hour urine sample, corresponding to the upper and lower limits of 1+, 2+, 3+ proteinuria in dipstick analysis and nephrotic range proteinuria, respectively. The limits of agreement between the two parameters were analysed by the Bland-Altman Plot, using the Med Calc statistical software version 7.6.0. This method depicts the mean difference and 95% confidence interval of the difference and limits agreement as mean difference ± 1.96 SD.

Results

The mean age of the study group was 74.9 (± 6.795) (minimum 65, maximum 91) years. Hypertension (HT) was detected in 92% (n=46), chronic kidney disease (CKD) in 70% (n=35), diabetes mellitus (DM) in 58% (n=29) and coronary arterial disease (CAD) in 46% (n=23) of the patients. The mean serum creatinine level was 1.31 g/dl (0.60-5.18), and the GFR was 42.50 ml/min/1.73 m² (minimum 8, maximum 103) (Table 1).

A strong and positive correlation was found between 24-HUP and PCR-SU ($r=0.879$, $p < 0.005$) (Figure 1). Although, there was a significant correlation between the two methods in all three groups, GFR < 30 ml/min/1.73 m² had the strongest correlation ($r=0.937$, $p < 0.005$). Correlation values decreased as GFR increased (GFR between 30-60 ml/min/1.73 m²: $r=0.801$, $p < 0.005$ and GFR > 60 ml/min/1.73 m²: $r=0.635$, $p < 0.005$). The ROC analyses, detected PCR-US discriminant values of 0.545, 0.465, 0.812, 3.683 mg/mg as indicators of ≥ 0.3 , ≥ 0.5 , ≥ 1.0 and ≥ 3.5 g/day of proteinuria in 24-HUP, respectively (Table 2). When the proteinuria level increased to ≥ 3.5 g/day, the sensitivity, specificity and discriminant values of PCR-SU also increased.

Discussion

This study detected a strong correlation between PCR-SU and 24-HUP in elderly patients with underlying HT, DM, CKD and CAD.

The world population is ageing. According to the World Health Organisation (WHO), first time in history, most people are expected to live over their sixties. Therefore, any medical laboratory test should be evaluated for coherence to use in the elderly. HT and DM are the most common chronic diseases in elderlies, and the main complication of these two diseases are kidney damage (10, 11). Furthermore, CKD and CAD incidence is

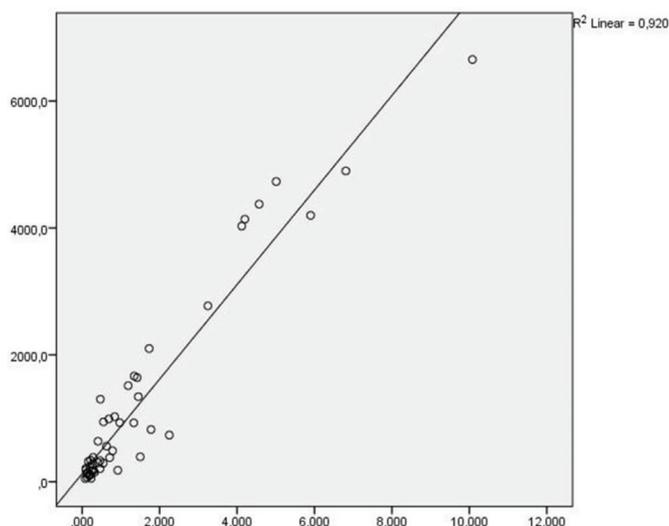


Figure 1. Correlation between spot urine protein-to-creatinine ratio and total protein level in 24-hour urine sample of the elderlies

increasing in this age group (12). So, it is evident that any test should be quick, cheap and reliable. In this regard, proteinuria detection in a urine sample is essential since both elderlies and disease burden with renal complications also increase. In our study, we demonstrated that proteinuria detection in spot urine is a reliable, sensitive and specific method. We found 80–85% sensitivity levels and 81–100% specificity levels of proteinuria detection with PCR-SU in elderlies. Our results are compatible with studies conducted on younger-aged populations (3, 6). Studies analysing the correlation between 24-HUP and PCR SU are frequently disease-specific and have younger populations (3, 4, 13).

According to our results, PCR-US is a convenient method for screening, diagnosis and follow-up of proteinuria in the elderlies. This correlation seems to be true for specific diseases and most common chronic conditions with renal complications in this age group.

The National Kidney Foundation/ Kidney Disease Outcomes Quality Initiative guidelines support the use of PCR-SU (preferably in the first morning urine or in the spot urine sample

at any hour if there is no first morning urine sample) to detect and monitor proteinuria (14). In our study, we collected our specimens as the first urine in the morning. Since our patients were hospitalised, we could also observe the patients' specimen collection appropriateness. This may be among the reasons of our higher specificity and sensitivity levels. Other studies are conducted mostly on outpatients (3, 6, 15, 16).

Various studies reported that 10–20% of the patients who collect 24-hour urine cannot follow the procedures of urine collection (16, 17). Due to cognitive losses, physical barriers and social problems in elderlies, it is more challenging to perform the examination correctly. Improper sample collection rates are, therefore, estimated to be much higher in the elderly. Not surprisingly, in our study, we detected that 31% (23) of the patients collected their urine samples inappropriately. Although our rates are slightly higher than reported in the literature, this may be because our patients were older, had geriatric syndromes impeding proper collection, and all were in-patients with acute problems. We also excluded patients with urinary incontinence, which has up to 30% prevalence and is among the main problems in collecting 24-hour urine in this age group. Therefore, we estimate that inappropriate specimen collection might be more frequent among the geriatric age group. From this point of view, we believe that proteinuria detection by PCR-SU in the elderly has more significant importance when compared with younger groups as 24-hour urine collection has higher rates of inappropriateness in the elderly.

Price et al. in their review of 16 articles comparing 24-HUP and PCR-SU for detection of proteinuria recorded that PCR-SU has a 69–96 % sensitivity and a 41–98% specificity for the detection of proteinuria of ≥ 300 mg/day. In this review, it was stated that evaluating proteinuria by the PCR-SU method may rule out the presence of significant proteinuria (18). In our study, by using the ROC curve and taking 24-HUP as the gold- standard method for proteinuria detection, we calculated the discriminant values of PCR-SU as 0.545 mg/mg, 0.465 mg/mg, 0.812 mg/mg and 3.683 mg/mg for ≥ 0.3 , ≥ 0.5 , ≥ 1.0 , and ≥ 3.5 g/day proteinuria

Table 1. Clinical and laboratory findings of the study population

Variable	
Male/female, percentage (number)	54/46% (27/23)
Age, years (mean±standard deviation)	74.9 ± 6.7
Hypertension, percentage (number)	92% (46)
Diabetes mellitus, percentage (number)	58% (29)
Coronary Arterial Disease, percentage (number)	46% (23)
Chronic Renal Disease, percentage (number)	70% (35)
Serum creatinine (gr/dl) (mean, min.-max.)	1.31 (0.60–5.18)
Glomerular Filtration Rate (MDRD) (ml/min/1.73m ²) (mean, min.-max.)	42.50 (8–103)
24-Hour urine protein mg/day (mean, min.-max.)	388 (50–6655)
Spot urine protein/creatinine ratio (mean, min.-max.)	0.545 (0.77–10.080)

Table 2. Discriminant protein- to-creatinine levels that predict threshold levels for proteinuria at 0.3, 0.5, 1.0 and 3.5 g/day

24-h urine total protein Threshold (g/day)	Discriminant values of spot urine PCR (mg/mg)	Sensitivity (95% CI)	Specificity (95% CI)	Area under the ROC curve (95% CI)
≥ 0.3	0.545	80.0 (62.69–90.49)	95.0 (76.39–99.11)	0.927 (0.856–0.998)
≥ 0.5	0.465	95.6 (79.01–99.23)	81.4 (63.30–91.82)	0.948 (0.892–1.0)
≥ 1.0	0.812	93.3 (70.18–98.81)	82.8 (67.32–91.90)	0.937 (0.873–1.0)
≥ 3.5	3.683	87.5 (52.91–97.76)	100.0 (91.80–100.00)	1.0 (1.0–1.0)

in 24-HUP with 80%, 95%, 93% and 87% sensitivity, and 95%, 81%, 82% and 100% specificity levels, all respectively. According to our results, we can say that PCR-SU has a high correlation and agreement with 24-HUP in elderly patients with different chronic diseases. Besides, when the patients were grouped according to GFR levels, there was a significant correlation between both methods in all groups, but GFR levels of <30 ml/min/m² was detected to have the highest correlation. In clinical practice, this will help clinicians since follow-up frequency increases as CKD progresses and more specific results are essential for clinical decision making of patients with advanced clinical diseases.

In summary, the main aim of geriatrics is comprehensive assessment of patients in a single centre with a multidisciplinary approach (19). Diminishing application rates to the hospital is among the aims of geriatric approach, especially for patients with transfer and mobilization difficulties. This study detected that PCR-SU is a highly sensitive and specific method with a high agreement with the gold-standard method. Since HT prevalence is up to 50-75%, and DM prevalence is up to 30% in the elderly, at least three out of four patients admitting to geriatrics clinics will be analysed for proteinuria, which is among complications of these and many other chronic diseases (10, 20, 21). Using PCR-SU instead of 24-HUP will decrease caregiver burden, patient burden as well as healthcare utilisation and health personnel burden. This is essential in the old-aged group, especially in those with limited mobility and transfer options, both economically, medically and socially. According to our results, we consider that PCR-SU can be used for proteinuria detection in elderly patients with chronic diseases affecting renal function, both for diagnosis and follow-up reliably. Malnutrition is a common geriatric syndrome, and proteinuria may alert clinicians for the planning of nutrition (22).

This study has some limitations and strengths. First, our sample size was limited. Thus, specific analyses for patients with different chronic diseases could not be performed. On the other side, to our knowledge this is the only study conducted in the elderly. Second, since our study population were in-patients, we can not generalise our findings for all patients. However, we were able to monitor the in-patients for appropriateness of sample collections. Furthermore, the same physician followed-up the specimens collection process.

In conclusion, PCR-SU is a highly sensitive and specific test with a high agreement with the gold-standard method for proteinuria diagnosis and follow-up in elderly patients with chronic diseases. This will help clinicians for clinical decision making in elderly patients, especially when they are frail, have restricted mobility or have difficulty in transfer secondary to medical, social or economic reasons.

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Ethics

Peer-review: Externally peer-reviewed.

Conflict of Interest: No conflict of interest was declared by the authors.

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References

- Barnas U, Schmidt A, Haas M, Kaider A, Tillawi S, Wamser P, et al. Parameters associated with chronic renal transplant failure. *Nephrol Dial Transplant*. 1997;12 Suppl 2:82-5.
- Hofmann W, Guder WG. A diagnostic programme for quantitative analysis of proteinuria. *J Clin Chem Clin Biochem*. 1989;27(9):589-600.
- Wahbeh AM, Ewais MH, Elsharif ME. Comparison of 24-hour urinary protein and protein:creatinine ratio in the assessment of proteinuria. *Saudi J Kidney Dis Transpl*. 2009;20(3):443-7.
- Natasha J, McIntyre MWT. How to measure proteinuria? . *Current Opinion in Nephrology and Hypertension*. 2008;17:600-3.
- Chen CF, Yang WC, Yang CY, Li SY, Ou SM, Chen YT, et al. Urinary protein/creatinine ratio weighted by estimated urinary creatinine improves the accuracy of predicting daily proteinuria. *Am J Med Sci*. 2015;349(6):477-87.
- Wahbeh AM. Spot urine protein-to-creatinine ratio compared with 24-hour urinary protein in patients with kidney transplant. *Exp Clin Transplant*. 2014;12(4):300-3.
- Antunes VV, Veronese FJ, Morales JV. Diagnostic accuracy of the protein/creatinine ratio in urine samples to estimate 24-h proteinuria in patients with primary glomerulopathies: a longitudinal study. *Nephrol Dial Transplant*. 2008;23(7):2242-6.
- Robert M, Sepandj F, Liston RM, Dooley KC. Random protein-creatinine ratio for the quantitation of proteinuria in pregnancy. *Obstet Gynecol*. 1997;90(6):893-5.
- Rodby RA, Rohde RD, Sharon Z, Pohl MA, Bain RP, Lewis EJ. The urine protein to creatinine ratio as a predictor of 24-hour urine protein excretion in type 1 diabetic patients with nephropathy. The Collaborative Study Group. *Am J Kidney Dis*. 1995;26(6):904-9.
- Zhao C, Wong L, Zhu Q, Yang H. Prevalence and correlates of chronic diseases in an elderly population: A community-based survey in Haikou. *PLoS One*. 2018;13(6):e0199006.
- Canlar S, Cinel M. In: Varlı M, editor. Yařlılarda Sık Görülen Durumlar 2: kronik hastalıklar. Ankara: Hedef CS Basın yayını; 2018.
- McClure M, Jorna T, Wilkinson L, Taylor J. Elderly patients with chronic kidney disease: do they really need a referral to the nephrology clinic? *Clin Kidney J*. 2017;10(5):698-702.
- Sanchez-Ramos L, Gillen G, Zamora J, Stenyakina A, Kaunitz AM. The protein-to-creatinine ratio for the prediction of significant proteinuria in patients at risk for preeclampsia: a meta-analysis. *Ann Clin Lab Sci*. 2013;43(2):211-20.
- Keane WF, Eknoyan G. Proteinuria, albuminuria, risk, assessment, detection, elimination (PARADE): a position paper of the National Kidney Foundation. *Am J Kidney Dis*. 1999;33(5):1004-10.
- Kayatas S, Erdogdu E, Cakar E, Yilmazer V, Arinkan SA, Dayicioğlu VE. Comparison of 24-hour urinary protein and protein-to-creatinine

- ratio in women with preeclampsia. *Eur J Obstet Gynecol Reprod Biol.* 2013;170(2):368-71.
16. Mitchell SC, Sheldon TA, Shaw AB. Quantification of proteinuria: a re-evaluation of the protein/creatinine ratio for elderly subjects. *Age Ageing.* 1993;22(6):443-9.
 17. Chitalia VC, Kothari J, Wells EJ, Livesey JH, Robson RA, Searle M, et al. Cost-benefit analysis and prediction of 24-hour proteinuria from the spot urine protein-creatinine ratio. *Clin Nephrol.* 2001;55(6):436-47.
 18. Price CP, Newall RG, Boyd JC. Use of protein:creatinine ratio measurements on random urine samples for prediction of significant proteinuria: a systematic review. *Clin Chem.* 2005;51(9):1577-86.
 19. Atmış V, Bahşı R, Öztörün HS, Coşardereioğlu Ç, Yalçın A, Aras S, Varlı M, Sürmeli DM, Turgut T. Public Awareness of Geriatrics in the 50th Year of Geriatrics in Turkey. *Eur J Geriatr Gerontol.* 2019;1:24-8.
 20. Gümüşsoy M, Bahşı R, Sürmeli DM, Turgut T, Öztörün HS, Atmış V, Varlı M, Aras S. [Insulin Misusage And Affect of Insulin Education in the Elderly]. *Van Medical J.* 2018;25:323-31.
 21. Canlar S, Güllü S. [Chronic complications of diabetes]. In: Vedia Tonyukuk Gedik ÖD, editor. *Clinical Endocrinology* 2017.
 22. Gümüşsoy M. [Nutrition in cancer patients. Current topics in hematooncology]. 2019. ed: Fatih Köse, Ahmed Kürşat Güneş, Ali Murat Sedef. Chapter 30. Ankara/Akademisyen Yayınevi.