GLOMERULAR FILTRATION RATE IN EXAMINED POPULATION OF BOSNIAN POSAVINA - REGION OF BALKAN ENDEMIC NEPHROPATHY

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Abstract

Balkan endemic nephropathy (BEN) is chronic tubulointersticial nephritis of unknown aetiology characterized by an insidious onset and gradual progression to end stage renal disease (ESRD). Endemic regions of Bosnia and Herzegovina are Posavina and Semberija, sited at basin of Sava River. In BEN, just like in other chronic renal diseases (CKD), glomerular filtration rate (GFR), is assumed a marker of overall renal function.

The aim of this study was to compare GFR in examinees of endemic and non-endemic region for BEN, and between examinees with and without risk factors for BEN within endemic region

Study included 603 inhabitants of Bosnian Posavina, out of whom 386 (65%) from endemic (Domaljevac) and 217 (36%) from non-endemic (Svilaj) village, and it was performed in two phases. The first phase encompassed obtaining anamnestic data (demographic, personal and family history), measurement of arterial blood pressure, and urine dipstick testing (specific gravity, pH, proteins, leukocytes, glucose, ketones, and mycroalbuminuria). In the second phase, besides repeated urine dipstick test, laboratory blood testing and abdominal ultrasound, with special attention to urinary tract, was also performed. We have compared GFR between examinees of endemic and non-endemic regions for BEN, and between examinees with and without family burden for BEN within endemic region, using MDRD formula for calculating GFR, with cut-off value (5th percentile) based on result of studies performed in European Caucasians in screening for CKD and for establishing stages of CKD in BEN. Medical was used for statistical testing.

Out of total number of examined inhabitants (603), 145 examinees were included in the second phase. After exclusion of 17 diabetic patients, 94 (73%) examinees from endemic and 34 (27%) examinees from non-endemic region remained. In the endemic region there were 46 (49%) examinees with and 48 (51%) without family burden for BEN. Overall GFR in examined groups was within physiologic range. There was not statistically significant difference in calculated GFR between examinees of endemic and non-endemic regions for BEN (Mann-Whitney test p=0.104; Fisher's test p=1), neither between examinees with and without family burden for BEN within endemic region (Mann-Whitney test p=0,7393; Fisher's test p=0,263).

Overall GFR in examined groups was within physiologic range. There wasn't statistically significant difference in calculated GFR between examinees of endemic and non-endemic regions for BEN, neither between examinees with and without family burden for BEN within endemic region. GFR, no matter how accurately calculated and estimated, does not represent significant biomarker for diagnosis, especially early diagnosis, of BEN, until maybe its overt advanced form.

KEY WORDS: BEN, Bosnia, endemic, non-endemic, family burden, GFR

Introduction

Balkan endemic nephropathy (BEN) is chronic tubulointersticial nephritis of unknown etiology characterized by an insidious onset and gradual progression to end stage renal disease with endemic regions sited at the basin of Danube and Sava Rivers, in more Balkan countries, including Bosnia and Herzegovina, more precisely in Posavina and Semberija. There were no changes in geographic and epidemiological characteristics of the disease since the first disease descriptions (1). Precise information exists on which regions and villages are endemic and which are spared, but sporadic cases of BEN in non-endemic regions are also mentioned in literature (2). Today, BEN is usually diagnosed in adult population, between 50 and 70, with slight predomination in female patients. Disease is characterized by frequent occurrence of tumors of renal pelvis and urethers.

Even tough numerous dilemmas and conflicting opinions regarding BEN etiology are encountered in literature, prevailing theory is that BEN is caused by chronic poisoning with aristolochic acid ingested by food in people with genetic predisposition to this disease. The dA-aristolactam and dG-AL DNA adducts are isolated in renal cortex of BEN patients, but not in patients with other chronic renal diseases (3). Therefore, BEN is considered a toxic tubulointerstitial nephropathy, with clinical picture and disease progression not differing from other tubulointerstitial nephropathies (1). For now, there are no specific biomarkers for diagnosis of BEN, so one is considered to have BEN if certain diagnostic criterions are met, and after exclusion of other kidney diseases. Internationally accepted criteria for diagnosing and classification of chronic kidney diseases (CKD) are used (4), among others, Modification of Diet in Renal Disease (MDRD) formula for estimating glomerular filtration rate (GFR), a common marker of overall renal function in CKD of different etiological background (5).

The aim of this study was to compare GFR in examinees of endemic and non-endemic region for BEN, and between examinees with and without family burden for BEN within endemic region.

Materials and Methods

Study included 603 inhabitants of Bosnian Posavina, out of whom 386 (65%) from endemic (Domaljevac) and 217 (36%) from non-endemic (Svilaj) village, and it was performed in two phases. The first phase encompassed obtaining anamnestic data (demographic, personal and family history), measurement of arterial blood pressure, and urine dipstick testing (specific gravity, pH, proteins, leukocytes, glucose, ketones, and mycroalbuminuria). In the second phase, besides repeated urine dipstick test, we also performed laboratory blood testing and abdominal ultrasound, with special attention to urinary tract. Combur-Test® Strips (Roche) and Micral-Test® strips (Roche) were used for those purposes. We have compared GFR between two groups: examinees of endemic and non-endemic regions for BEN, and between two subgroups: examinees with and without family burden for BEN (offspring of families with history of BEN) within endemic region, using MDRD formula for estimating GFR, with cut-off value (5th percentile) based on results of studies performed in European Caucasians in screening for CKD and for establishing stages of CKD in BEN. Biostatistical programme MedCalc was used for statistical testing.

Results

Out of total number of examined inhabitants (603), 145 (24%) examinees were included in the second phase. After exclusion of 17 diabetic patients, 94 (73%) examinees from endemic and 34 (27%) examinees from non-endemic region remained, out of whom 12 male (35 %) and 22 female (65 %). In the endemic region there were 46 (49%) examinees, 21 (47 %) male and 25 (54 %) female, with and 48 (51%), 18 (38 %) and 30 (63 %) female, without family burden for BEN. Overall GFR in examined groups was within physiologic range. There was not statistically significant difference in calculated GFR between examinees of endemic and non-endemic regions for BEN (Mann-Whitney test p=0,104; Fisher's test p=1), neither between examinees with and without family burden for BEN within endemic region (Mann-Whitney test p=0,7393; Fisher's test p=0,263). Table 1 shows average age and median of estimated GFR for

different groups of examinees. There were 16 (13 %) of those with GFR below 60 ml/min, 6 (38%) men and 10 (63 %) women, and 82 (64 %) of those with GFR below 90 ml/min, 31 (38 %) men and 51 (62 %) women. Table 2. shows distribution of examinees according to value of estimated GFR. When compared to cut-off values (5th percentiles) from BEN consensus, low GFR was found in only 12 (9 %) examinees (Table 3). GFR above 120 ml/min was found in only two examinees (2%).

Group (n)	Average age X ± SD	GFR median (95 % CI)
Endemic village examinees with family burden (46)	49,87 ± 14,57	84,4 (72,9-93,3)
Endemic village examinees without family burden (48)	54,06 ± 15,98	86,0 (81,4-90,7)
Examinees from non-endemic village (34)	59,41 ± 15,7	80,3 (67,2-87,3)

GFR – glomerular filtration rate BEN – Balkan endemic nephropathy

TABLE 1. Average age and GFR medians in examinees of Bosanska Posavina

Group (n)	GFR < 60 ml/min	GFR < 90ml/min
Endemic village examinees with family burden (46)	7/46 (15%)	27/46 (59%)
Endemic village examinees without family burden (48)	4/48 (8%)	28/48 (58%)
Total (endemic village)	11/94 (11%)	55/94 (59%)
Examinees from non-endemic village (34)	5/34 (15%)	27/34 (79%)
Total (128)	16/128 (12,5%)	82/128 (64%)

GFR – glomerular filtration rate
BEN – Balkan endemic nephropathy

TABLE 2. Distribution of examinees according to GFR

Group (n)	M	F	total (%)
Endemic village examinees with family burden (46)	2/21	3/25	5/46 (10,9%)
Endemic village examinees without family burden (48)	0/18	2/30	2/48 (4,2%)
Examinees from non-endemic village (34)	2/12	3/22	5/34 (14,7%)
Total (128)	4/51	8/77	12/128 (9,4%)

GFR – glomerular filtration rate BEN – Balkan endemic nephropathy
M- male; F- female

TABLE 3. Examinees with decreased GFR according to 5th percentile

Discussion

We have conveyed this study in the region of Bosanska Posavina, with known endemic BEN regions, following recommendations given by the KDIGO for testing high risk groups for CKD. Because of the older age of individuals at the onset of many kidney diseases (BEN in particular, now between 50 and 70), the slow rate of decline of kidney function and high death rate due to CVD, most individuals with CKD do not develop kidney failure. However, decreased GFR is associated with a wide range of complications, such as hypertension, anaemia, malnutrition, bone disease, neuropathy, and decreased quality of life. Therapeutic interventions at earlier stages can prevent or ameliorate most of the complications of decreased kidney function, as well as slow the progression to kidney failure. Thus, measures to improve prevention, detection, and treatment of CKD in these earlier stages could reduce adverse outcomes and improve the quality of life of individuals with CKD (4) Bearing in mind that in the second phase of our study the examined population as a whole had some kind of kidney disease in a form of kidney damage, since they had albuminuria in two examined first morning urine samples, we proceeded with determining if they had lower GFR, that would put them in the higher risk group for developing overt CKD and other complications. We put emphasis on differences between BEN endemic and non-endemic regions, and between patients with and those without BEN family burden within endemic region. We have tested differences in estimated GFR among mentioned groups of patients, by using different criterion for estimation. When criterion suggested in BEN consensus (6, 7) was used, the frequency of examinees with low GFR was the lowest, even lower than when threshold level of < 60 ml/min/1,73 m2 was used, as proposed by KDIGO (4). Irrespective of the mentioned criterions, we did not found significant difference in frequency of low GFR between endemic and non-endemic regions, neither between those with and without BEN burden within endemic region. Difference was not significant in GFR median among tested groups also. Mentioned findings are similar with those found in older and recent literature dealing with same dilemma. In early 90tees, Vukusic et al. studied GFR in examinees without manifestations of kidney disease, in BEN endemic regions of Serbia. Sample was small, with 40 examinees from endemic region with family burden for BEN and 10 also from endemic region, but without family burden. GFR was calculated using diethylenetriamine pentaacetic acid (DTPA) clearance and it was within physiologic range in the whole sample, without statistical significant difference regardless of family burden for BEN. In the same study, authors were also performing other functional tests in order to estimate renal function, which in turn led them to the conclusion that those with family burden for BEN had preserved global and partial renal function with compensatory adaptation of functioning nefrons (exaggerated natriuresis, excretion fraction of sodium on the upper limit

of normal values, high maximal urine osmolality) (8). Meanwhile, on the sample of 93 healthy examinees from endemic families no changes in GFR were found (9). Radonic in 1992 also describes changes in urine concentration ability, occurring some time before decease in renal blood flow and GFR (10). Dimitrov et al in 2004 examined renal clinical markers in offspring of endemic families (131 examinees) and offspring of non-endemic families (102 examinees) from Bulgary. They revealed that GFR was within physiologic range in healthy offspring of endemic families, with 88,9 ml/min in examinees suspicious of developing the disease and 85 ml/min in examinees with BEN manifestations (11). Glomerular filtration rate median of examinees from endemic region in our study was similar to that found in study of Dimitrov et al for examinees with BEN manifestations, but still, only 7% of our examinees from endemic region were assumed as having decreased GFR compared to their cut-off (5th percentile) value. Hence, if GFR of BEN patients from Dimitrov's study were subsequently compared to 5th percentile for certain sex and age, now offered in new BEN Consensus Statement, we may found that even those BEN patients had normal values of GFR. In study conveyed in Serbia on 98 examinees, offspring of endemic families, 25 immigrant in endemic families, and 50 of those without BEN burden. Authors didn't confirm any statistically significant difference in GFR between examined groups (12). Stasevic et al conveyed screening of inhabitants of endemic region of Vitina. Overall 510 persons were examined and their GFR estimated by using Cockroft-Gaoult equation. In this study 46,5% of inhabitants had GFR bellow 100 ml/min, while 12% had GFR bellow 60 ml/min. They also registered 13% of those with GFR above 140 ml/min, among whom 12 examinees with GFR above 180 ml/min. Authors assumed that this speaks in favour of hiperfiltration in early stages of BEN (13). Similarly, in our study, in endemic village there were 59 % of those with GFR bellow 90 ml/min and 11 % of those with GFR bellow 60 ml/min. On the other hand, there were a substantial percentage of examinees with GFR < 90 ml/min/1,73 m2, with significantly higher frequency in group from non-endemic region. This can be partially explained by the fact that this group had the highest average age of examinees, since GFR lowers with aging, together with the fact that other diseases influencing GFR progress with time. There were only 2 % with GFR above 120 ml/min. Hyperfiltration in early stages of BEN was also described by Djukanovic et al among renal patients with suspected BEN. Authors noticed that kidney length in patients with GFR above 120 ml/min was significantly lower in patients with BEN compared to patients with other primary kidney diseases (14). In one small study 18 examinees, with pathohystologically confirmed diagnosis of BEN 15 years prior to study, were followed. The observation was that GFR was declining by average of 2,74 ml/min per year, but in two patients increment in GFR was noticed (15). In Bosnia and Herzegovina the GFR of BEN patients was tested in late 90tiees of twentieth century. There were 59 patients with pathohystologically confirmed diagnosis. Authors concluded that GFR, estimated by DTPA clearance, was directly proportional to kidney volume, but that average kidney volume was bellow referent borderline values in all patients, even in the group (11 patients) with average GFR of 120,65 ml/min (16). This observation that individuals with BEN may have hyperfiltration rendering higher GFR, in our opinion is not sufficiently exploited and explained, and our study did not give us enough evidence to prove or contribute to aforementioned theory, but interestingly those examinees were from the endemic village. In the majority of the studies that we have encountered while going through the literature of relevance for this study, investigators have used DTPA for GFR determination or Cockroft-Gault equation for calculating it. In our study, we have calculated GFR using more accurate MDRD formula. According to KDIGO guidelines (17, 4) GFR should be estimated utilizing MDRD formula, while Cockroft-Gault equation proved less accurate in healthy subjects than in patients with CKD. It is also noticed that this equation offers less accurate information in elderly, and also overestimates GFR when renal function is seriously impaired. Likewise, MDRD equation has some limitations, for example it underestimates GFR in individuals with normal serum creatinine levels, thus, may erroneously categorize certain healthy subjects as having CKD. Hence, it is advisable to report exact GFR value, and not the stage of CKD, especially in case of estimated GFR < 60 ml/min. Also, usage of age and gender-specific lower limits of normal GFR values are proposed and tables with reference values of GFR in a European Caucasians population are offered (6). Another important advantage of our study is that we have minimized the chance of erroneous estimation of GFR by using the cut-off value (5th percentile) based on result of studies performed in European Caucasians in screening for CKD and for establishing stages of CKD

in BEN. To our knowledge, previous similar studies did not use this cut-off value. By using that we have proved that there are less people considered as having low GFR than in case of using one universal cut-off value of < 60 ml/min/1,73 m2. Even when using this strict criterion, there were no significant differences between endemic and non-endemic region and between those with and without BEN burden in endemic region.

Conclusion

Overall GFR in examined groups was within physiologic range. There wasn't statistically significant difference in calculated GFR between examinees of endemic and non-endemic regions for BEN, neither between examinees with and without family burden for BEN within endemic region. GFR, no matter how accurately calculated and estimated, does not represent significant biomarker for diagnosis, especially early diagnosis, of BEN, until maybe its overt advanced form.

References

(1) Bamias G., Boletis J. Balkan Nephropathy: Evolution of Our Knowledge. AJKD 2008; 52: 606-616.

(2) Nikolić J. Epidemijska nefropatija i tumori gornjeg urotela. Sporadična endemska nefropatija. Izdavacko preduzece Beograd A.D. 2006; 171-181.

(3) Grollman A.P., Shibutani S., Moriya M., Miller F., Wu L., Moll U. et al. Aristolochic acid and the etiology of endemic (Balkan) nephropathy. PNAS 2007; 104: 12129-12134.

(4) Levey A., Eckardt K.U., Tsukamoto Y., Levin A., Coresh J., Rosert J., De Zeeuw D., Hostetter T., Lamiere N., Eknoyan G. Definition and classification of chronic kidney disease: A position statement from Kidney Disease: Improving Global Outcomes (KDIGO). Kidney Int. 2005; 67: 2089-2100.

(5) Levey S., Atkins R., Coresh J., Cohen E.P., Collins A.J., Eckardt U.K. et al. Chronic kidney disease as a global public health problem: Approaches and initiatives – a position statement from Kidney Disease Improving Global Outcomes. Kidney Int. 2007; 72: 247-259.

(6) Wetzels J., Kiemeney L., Swinkels D., Willems H., den Heijer M. Age and gender specific reference values of estimated GFR in Caucasians: The Nijmegen Biomedical Study. Kidney Int. 2007; 72: 632-637.

(7) Anonymous. Consensus Statement. International Workshop on Screening, Diagnosis, Classification and Treatment of Endemic (Balkan) Nephropathy. Island of Brač, Croatia, April 2008.

(8) Vukušić Z., Radenković S., Tasić S., Trajkovski S., Raičević R. Funkcionalno stanje bubrega u ispitanika sa endemskog zarista bez klinicki manifestnog oboljenja. Etiology of endemic (Balkan) nephropathy. Proceedings of the 6th Symposium on endemic (Balkan) nephropathy, Niš 1987; 153-158.

(9) Bogićević M., Strahinjić S., Stefanović V. Protok plazme kroz bubreg i glomerulska filtracija u zdravih osoba iz porodica sa endemskom nefropatijom. Etiology of endemic (Balkan) nephropathy. Proceedings of the 6th Symposium on endemic (Balkan) nephropathy, Niš 1987, 159-163.

(10) Radonić M., Radošević Z. Clinical features of Balkan endemic nephropathy. Food Chem. Toxicol. 1992; 30: 189-192.

(11) Dimitrov P., Karmaus W., Simeonov V. Follow-up of BEN offspring in Vratza, Bulgaria and new incident cases. Recent advances in Balkan endemic nephropathy research. Srpska akademija nauka i umetnosti, Beograd 2009; p. 24.

(12) Stefanović V., Cukuranović R., Djordjević V., Jovanović I., Marinković D. A study of adult offspring from families with Balkan endemic nephropathy. Recent advances in Balkan endemic nephropathy research. Srpska akademija nauka i umetnosti, Beograd 2009, p 25.

(13) Stašević Z., Subaric-Gorgijeva G., Krčmarevic J., Stolić R., Trajković G. Results of kidney function measurement and blood pressure in population from endemic region of Vitina. Med. Pregl. 2008; 7-8: 401-403.

(14) Djukanović L., Bukvić D., Marić I., Cukuranović R., Vukomanovic M., Glogovac S. Open questions on Balkan nephropathy. Nephrol. Dial. Transplant. 2001; 16 (Suppl 6): 27-29.

(15) Cukuranović R., Savić V., Stefanović N., Stefanović V. Progression of kidney damage in Balkan endemic nephropathy: A 15-year follow up of patients with kidney biopsy examination. Renal Failure 2005; 27: 701-706.

(16) Trnačevic S., Halilbasić A., Ferluga D., Plavljanić D., Vizjak A., Duraković H., Habul V., Mesić E., Imamović G., Hranisavljević J., Pašić M., Paunović G. Renal function, protein excretion and pathology of Balkan endemic nephropathy. I Renal function. Kidney Int. 1991; 40 (Suppl 34): S49-S51.

(17) Anonymous. National Kidney Foundation: KDOQI Clinical Practice Guidelines for Chronic Kidney Disease: Evaluation, Classification and Stratification. AJKD 2002; 39 (Suppl 1): S1-S266.