



Evaluation of renal tubular function in epileptic children treated with levetiracetam

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Abstract

Our study aim was to measure the urinary N-acetyl- β -D-glucosaminidase/creatinine (NAG/UCr) index in epileptic children who received levetiracetam (LEV) treatment at least for 6 months, and compare it to healthy children. Thirty five children with epilepsy were enrolled in this prospective study. NAG was studied using the calorimetric method and NAG levels were expressed in units per liter (U/L) and NAG/UCr levels were determined in U/mmol creatinine. There were no statistically significant differences for the urine NAG and NAG/UCr index before and after LEV treatment in the epileptic group ($p > 0.05$, for each). There were no significant correlations between the serum concentration of LEV and urinary NAG levels ($r = 0.258$, $p = 0.135$) and NAG/UCr levels ($r = 0.164$, $p = 0.346$) before treatment. Our study demonstrated that LEV treatment was safe and did not interfere in renal tubular function in epileptic children.

Keywords: Children, epilepsy, levetiracetam, N-acetyl- β -D-glucosaminidase, renal tubular function

Introduction

Urinary N-acetyl- β -D-glucosaminidase (NAG), a hydrolytic enzyme, has two isoenzymes in humans. The NAG-A isoenzyme is present in the lysosomes of renal proximal tubular cells and its increased concentration in urine is considered as a marker of acute kidney injury [1-4].

Because of insufficient seizure control or important side effects during antiepileptic drugs (AEDs), epilepsy treatment in children is a difficult task in clinical practise [5,6]. Levetiracetam (LEV) is used for the treatment of partial-onset, myoclonic and primary generalized tonic-clonic seizures in children with a minimal side effect profile, not only as an adjunctive therapy but also as single-agent therapy for epilepsy [7]. AEDs may lead to impairment in renal function after a long treatment period [8-12]. While some studies have disputed the urinary NAG as a marker of AEDs-induced renal functional impairment [13-16], some others disclosed that the increased excretion of tubular enzymes and proteins does not necessarily owe to the side-effects of the AEDs but is most likely due to a physiological change in renal function that is related to epilepsy itself [17]. Thus, it is not clear whether epilepsy itself or the AEDs cause kidney damage. To the best of our

knowledge, there are only two case reports about NAG levels in epilepsy patients treated with LEV, however, there are no further studies in the literature [11,12]. Therefore, in our study it was planned to evaluate the effects of LEV treatment on renal function in epileptic children. For that purpose we aimed to measure NAG/urine creatinine (UCr) activity index in epileptic children who received LEV treatment at least for 6 months and compare it to healthy children.

Material and Methods

Study population

Thirty five children with epilepsy, who were followed by the department of pediatric neurology, were enrolled in this prospective study. Eighteen patients were boys and 17 were girls. Their median age was 8.0 years (5.0–15.5 yr.). Thirty five healthy children were included as controls. Of them, 14 (40%) were boys and 21 (60%) were girls and their median age was 8.5 years (1.0–17.0). None of the healthy children were on any medication at the time of urine sampling. All patients were ambulatory and none had mental retardation or neurological abnormalities. Children with the signs of renal dysfunction or those taking nephrotoxic medication and those with a previous history of renal disease, diabetes mellitus, liver diseases, chronic, metabolic, systemic or neurodegenerative diseases were excluded from the study. Before medical therapy was initiated, blood urea nitrogen (BUN) (mg/dl), serum creatinine (SCr) (mg/dl), serum uric acid (mg/dl), liver

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function tests [**aspartate aminotransferase** (AST)(U/L) and alanine aminotransferase (ALT) (U/L)] in the blood were evaluated. In addition, spot complete urine analysis for protein and glucose, UCr, urine sodium (Na) and urine NAG levels were measured. After this procedure, all patients with epilepsy received LEV treatment at least for 6 months. After 6-12 months of LEV treatment, all measures were repeated in the epileptic patients. The patients did not have any sign of renal dysfunction. BUN, SCr, uric acid and, liver function tests (AST and ALT) were in the normal range in all patients. None had received aminoglycoside or other nephrotoxic treatment since epilepsy was diagnosed. Of the 35 patients with epilepsy, 22 had focal seizures, 6 had generalized tonic-clonic seizures, 5 had juvenile myoclonic epilepsy, one had complex partial epilepsy, and one had startle type epilepsy. The seizures were well controlled in these patients after the AEDs had been adjusted to therapeutic levels. The LEV doses given to the patients were a minimum of 10 mg/kg/day, and a maximum of 60 mg/kg/day.

The procedures were performed according to the Declaration of Helsinki for the ethical standard for human experimentations. The study was approved by the Ethics Committee of Inonu University and written consent forms were signed by the parents before participating in the study.

NAG evaluation

Biochemical parameters such as BUN, SCr, uric acid, AST and ALT levels, and urinary parameters including urine sodium and UCr were immediately studied. For the NAG measurement, the urine sample was centrifuged at 4,000 rpm for 8 min and supernatant was stored at -70°C until the NAG measurement was achieved, according to the manufacturer's protocol. NAG was studied using the calorimetric method with Diazyme kit at 505-nm wave length calibrator (Shimadzu UV-1201V Spectrophotometer, Siemens, Columbia, USA). NAG levels were expressed in units per liter (mIU/L) and NAG/UCr levels were determined in mIU/mg creatinine.

Statistical analysis

The statistical analysis of this study was performed using the Statistical Package for Social Sciences program (SPSS) for Windows version 16.0 (SPSS Inc., Chicago, Illinois, United States). Numerical data are expressed as

mean $\pm$ standard deviation or median (with minimum-maximum range), depending on normal distribution (Shapiro-Wilk test). While differences between the two groups were assessed by using unpaired t test or Mann-Whitney U test, and the differences between the two groups were assessed by using paired t test or the Wilcoxon signed rank test. Categorical variables in proportions or percentages were analyzed by the chi-square test or the Fisher exact test when appropriate. Associations between variables were assessed by Spearman's correlations analysis. A p-value less than 0.05 was considered statistically significant.

Results

There were no differences in the male to female ratio as well as median age, weight and height between the patients and controls ($p=0.472$, $p=0.962$, $p=0.668$, and $p=0.771$, respectively). All patients had normal results on urinalysis, serum BUN, SCr, serum uric acid, AST, and ALT levels. No statistically significant differences were found between patients and controls with respect to BUN, SCr, serum uric acid, AST, or ALT levels ($p>0.05$, for each). All urine samples of patients were negative for protein and glucose. In addition, there were no significant differences in urine NAG levels and NAG activity between the patient group and healthy group (Urine NAG levels 6.5 versus 11.0 and NAG index: 0.007 versus 0.008, $p=0.087$ and $p=0.202$, respectively). The clinical and laboratory data of the epileptic patients and controls were given in Table 1.

No statistically significant differences were found in BUN, SCr, serum uric acid, AST, and ALT levels ($p>0.05$, for each) of epileptic group before and after LEV treatment. In addition, there were no statistically significant differences for urine NAG and NAG/UCr index before and after LEV treatment ($p>0.05$, for each). The laboratory data of the epileptic patients before and after LEV treatment were shown in Table 2.

There were no significant correlations between the serum concentration of LEV and urinary NAG levels ($r=0.258$, $p=0.135$) and NAG/UCr levels ($r=0.164$, $p=0.346$) before treatment. Likewise, there were no significant correlations after treatment ($r=0.153$, $p=0.380$ for urinary NAG level, and $r=0.54$, $p=0.757$ for NAG/UCr excretion).

Table 1. The clinical and laboratory data of the epileptic patients and controls

Parameter	Epileptic group (n=35)	Control group (n=35)	p
Age (year)	10,2±3,7	10,2±4,8	0,945
Gender (M/F)	18/17	14/21	0,472
Weight (kg)	37,6±16,9	40,5±19,4	0,511
Height (cm)	139,2±19,6	140,0±26,4	0,886
BUN (mg/dl)	10,0±2,9	11,1±3,3	0,900
SCr (mg/dl)	0,52±0,07	0,51±0,09	0,665
Uric acid (mg/dl)	3,4±1,2	3,7±1,3	0,620
AST (U/L)	20,3±10,8	25,7±8,6	0,50
ALT (U/L)	18,7±9,0	16,4±7,2	0,301
Urine creatinine (mg/dl)	106,2±64,8	112,5±70,6	0,701
Urine NAG (mIU/L)	11,8±10,7	18,4±15,5	0,041
NAG/Cre index (mIU/mg creatinine)	0,17±0,24	0,25±0,30	0,268

M: Male, F: Female, BUN: Blood urea nitrogen, SCr: Serum creatinine, AST: *Aspartate aminotransferase*, ALT: Alanine aminotransferase.

Table 2. The laboratory data of the epileptic patients before and after levetiracetam treatment

Parameter	Before treatment (n=35)	After treatment (n=35)	p
BUN (mg/dl)	10,0±2,9	10,6±2,9	0,203
SCr (mg/dl)	0,52±0,07	0,52±0,07	0,858
Uric acid (mg/dl)	3,4±1,2	3,3±1,1	0,553
AST (U/L)	20,7±10,8	22,0±6,3	0,489
ALT (U/L)	18,8±9,1	14,5±9,1	0,075
Urine creatinine (mg/dl)	106,2±64,8	115,4±56,7	0,416
Urine NAG (mIU/L)	11,8±10,7	9,8±8,4	0,340
NAG/Cre index (mIU/mg creatinine)	0,17±0,24	0,11±0,14	0,167

BUN: Blood urea nitrogen, SCr: Serum creatinine, AST: *Aspartate aminotransferase*, ALT: Alanine aminotransferase.

Discussion

Our study could not confirm the data of previous reports that revealed an increased excretion of tubular NAG enzyme in children receiving AEDs treatment other than LEV. Our study showed that the median values of urine NAG and NAG/UCr index were not different before and after LEV treatment in epileptic children. Our study demonstrated that LEV treatment was safe and did not impede renal tubular function in epileptic children.

LEV that is constitutionally and chemically irrelevant to present AEDs includes a single enantiomer [18]. Studies demonstrated that LEV is most powerful for the treatment of complex partial epilepsy not with standing recent reports which display efficiency in the treatment of many seizure types in children [19,20]. LEV is digested orally, and its excretion from the body is entirely renal. Of the total LEV dose, approximately 34% is metabolized and 66% is excreted unmetabolized in the urine [21]. LEV is swiftly filtered by the kidneys, and greater than 90% of the LEV is excreted within 48 hours, depending on the glomerular filtration rate (GFR). It is filtered by the glomeruli and is dependent on partial tubular reabsorption [18,21]. When loading or titrating dosages of LEV in epilepsy patients, decreased GFR is an important indication for AEDs reduction [22]. In children, renal clearance is higher, and

dosage ought to be augmented to nearly 130% of the adult dose on a per kilogram of body weight basis [21].

Side effects of LEV treatment are somnolence, accidental injury, vomiting, anorexia, rhinitis asthenia, irritability, and behavior problems [23,24]. Renal toxicity related to LEV has been previously seen in only one child [25]. In a recent multicenter, randomized, placebo-controlled study, in which 198 children who have poorly controlled partial seizures were included, renal toxicity was not diagnosed [26]. However, only one case has been formerly noted to lead to significant renal complications during LEV treatment. The case of a formerly healthy 17-year old child with normal renal function, who afterwards developed interstitial nephritis and renal failure during administration of an appropriate dose of LEV was reported. AEDs including phenytoin, oxcarbamazepine, phenobarbital, and valproic acid have been reported to cause to allergic renal adverse effects [27,28]. An aromatic ring is considered being the structural common link in AEDs that brings about allergic interstitial nephritis [29]. LEV does not include an aromatic ring, though the potential risk for hypersensitivity renal complications is formerly unpredicted. In our study, consistent with previous studies, we did not observe renal toxicity secondary to LEV treatment. Before and after LEV treatment, all patients' renal function tests were detected to be within normal

range and LEV treatment did not impede renal function tests in children with epilepsy.

Some studies demonstrated that side effects of mono and combined AEDs on renal tubular function measured by NAG/UCr index, while the others described the clinical efficiencies of these drugs in epileptic children. In a previous study [14], it was revealed that the NAG levels were high in 29% of all patients, in 47% of a valproate group, and in 38% of a carbamazepine group. The mentioned study also showed that a significant positive correlation was observed between NAG/UCr index and serum concentration of valproate. Another study [15] revealed that VPA management was in line with the highest incidence of abnormal urinary NAG/UCr index. Nevertheless, the incidence of high urinary NAG/UCr index was significantly similar in the poly-therapy and mono-therapy groups. A previous study [30] demonstrated an increase in the NAG/UCr activity index associated with concomitant therapy, but that was in the normal limits. So, tubular dysfunction was not confirmed subsequent to their treatment plan. Another study depicted that those on AEDs treatment with therapeutic drug levels showed minor signs of tubular dysfunction [31]. Furthermore, NAG is a lysosomal hydrolases and it plays an important role in the catabolism of both glycoproteins and glycosaminoglycans [32]. This mechanism essentially takes place in the proximal tubulus and thus can be seen as a marker of functional impairment of tubules [31,32]. In our study, we did not observe side effects of renal function tests and the NAG/UCr index before and after LEV treatment.

In our study, the duration of LEV treatment in all epileptic patients was similar and this therapy continued for at least 6 months. After 6-12 months of LEV treatment no negative effect on renal function tests was observed. In addition, the NAG/UCr activity index was similar before and after LEV treatment, independent of therapy duration. Hence, we suggest that LEV treatment is a safe and credible drug in epileptic children, especially those with renal impairment.

Conclusion

Thus, according to our study results, the NAG/UCr index can be used as a sensitive indicator of renal tubular disease activity, and it might be a suitable screening test for early diagnosis of renal disturbance. However, our study demonstrated that LEV treatment was safe and did not impede renal tubular function in epileptic children. Therefore, LEV treatment may be safely used to treat renal function impairment in epileptic children.

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