Assessment of the renal function, peroxidation damage and inflammatory injury after epalrestat combined with alprostadil treatment of early diabetic nephropathy

Hai-Xia Li, Shu-Juan Liu

Health Care Department for Cadres, The First People’s Hospital of Xianyang City Shaanxi Province, Xianyang 712000, China

ARTICLE INFO

Article history:
Received 7 Jul 2016
Received in revised form 17 Jul 2016
Accepted 12 Jul 2016
Available online 24 Jul 2016

Keywords:
Early diabetic nephropathy
Epalrestat
Alprostadil
Renal function
Peroxidation damage

ABSTRACT

Objective: To study the renal function, peroxidation damage and inflammatory injury after epalrestat combined with alprostadil treatment of early diabetic nephropathy. Methods: 90 patients with early diabetic nephropathy treated in our hospital between June 2011 and November 2015 were collected and divided into observation group and control group (n=45) according to the single-blind randomized control method. Observation group received epalrestat combined with alprostadil treatment, control group received alprostadil treatment alone, and the treatment of both groups lasted for 3 months. Before treatment and after 3 months of treatment, turbidimetric immunoassay was used to detect the renal function indexes in peripheral blood, rate method was used to detect the renal function indexes in urine, and ELISA method was used to detect the levels of peroxidation indexes and inflammation indexes. Results: Before treatment, differences in renal function, peroxidation damage and inflammatory damage indexes were not statistically significant between two groups of patients (P>0.05). After 3 months of treatment, creatinine (Scr), cystatin C (CysC), β2 microglobulin (β2-MG), N-acetyl-β-D-glucosaminidase (NAG), reactive oxygen species (ROS), advanced protein oxidation products (AOPPs), interleukin-8 (IL-8), interleukin-27 (IL-27) and procalcitonin (PCT) levels of observation group were lower than those of control group while catalase (CAT), total superoxide dismutase (TSOD), interleukin-4 (IL-4), interleukin-10 (IL-10) and interleukin-13 (IL-13) levels were higher than those of control group (P<0.05). Conclusions: Epalrestat combined with alprostadil can protect the renal function and inhibit the peroxidation damage and inflammatory injury in patients with early diabetic nephropathy.

1. Introduction

Diabetic nephropathy is one of the most important organ complications in diabetic patients with a long course of disease and poor blood glucose control, it is without any abnormal results of laboratory tests in early stage, patients’ renal function declines gradually with the disease progression, and will finally progress to renal failure[1,2]. Early diabetic nephropathy is the special stage of diabetic nephropathy, also called "continuous microalbuminuria period", glomerular nodular changes occur in renal pathology of patients in this period, and the urine albumin excretion (UAE) rate rises to 20–200 μg/min and will continue to rise without active intervention. Many studies have shown that the renal abnormalities are still reversible in early stage of diabetic nephropathy, alprostadil is a commonly used drug to treat diabetes-related complications, it is with multiple effects such as improving hemodynamics and hemorheology and dilating vascular smooth muscle, it has been confirmed to be able to reduce the UAE of patients with diabetic nephropathy to a certain extent, but the curative effect has limitations[3]. Epalrestat belongs to aldose reductase inhibitor, can inhibit the polyol metabolism process and has been successfully applied in diabetic neuropathy, some scholars have proposed to use epalrestat in the adjuvant treatment of patients with early diabetic nephropathy, but there are not many studies on its role in protecting renal function and stabilizing internal environment[4]. In the study, epalrestat combined with alprostadil was used in the treatment of patients with early diabetic nephropathy, and the effect of epalrestat combined with alprostadil treatment on renal function, peroxidation damage and inflammatory injury in patients with early diabetic nephropathy was mainly analyzed.

[Corresponding author: Shu-Juan Liu, Health Care Department for Cadres, The First People’s Hospital of Xianyang City Shaanxi Province, Xianyang 712000, China. Tel: 14729041609
Fund Project: Scientific Research Projects of Xianyang Science and Technology Bureau [No. XK07018-(5)].]
2. Materials and methods

2.1. Inclusion and exclusion criteria

Inclusion criteria: (1) in accordance with the clinical diagnostic criteria for early diabetic nephropathy; (2) without hydrenephrosis, chronic glomerulonephritis, kidney neoplasms and other diseases; (3) with normal cognitive function and participating in the whole treatment. Exclusion criteria: (1) with serious heart and liver dysfunction; (2) allergic to epalrestat and (or) alprostadil; (3) associated with systemic infectious diseases; (4) with malignant tumor diseases; (5) with surgery history within 1 month prior to admission.

2.2. Clinical information

90 patients with early diabetic nephropathy treated in our hospital between June 2011 and November 2015 were included and divided into observation group and control group (n=45) according to the single-blind randomized control method. Control group included 24 male cases and 21 female cases, they were 50–78 years old, the course of diabetes was 5–19 and (12.18±4.76) years in average, and the course of diabetic nephropathy was 3 months–2 years and (8.82±1.61) months in average; observation group included 25 male cases and 20 female cases, they were 51–79 years old, the course of diabetes was 6221 and (13.59±4.82) years in average, and the course of diabetic nephropathy was 2 months22 years and (8.69±1.75) months in average. Two groups of patients were not statistically different in the distribution of gender, age, course of diabetes and course of diabetic nephropathy (P>0.05), the included patients themselves signed the informed consent, and the research process was approved by the hospital ethics committee.

2.3. Treatment methods

Both groups of patients received insulin for regular hypoglycemic therapy, and control group of patients accepted alprostadil treatment on the basis of conventional treatment, specifically as follows: intravenous injection of alprostadil (Wuhan Aimin Pharmaceutical Co., LTD., approved by H42022501) 10 μg±10 mL saline mixture, 1 time/d, for consecutive 3 months of treatment. Observation group of patients accepted epalrestat combined with alprostadil treatment on the basis of conventional treatment, specifically as follows: oral administration of epalrestat tablets (Yangtze River Pharmaceutical Group Nanjing Hailing Pharmaceutical Co., LTD., approved by H20040012), 50 mg/time, 1 time/d; alprostadil usage and dosage were the same as those of control group, and both groups lasted for 3 months.

2.4. Observation indexes

2.4.1. Renal function indexes

Before treatment and after 3 months of treatment, 2 mL of fasting peripheral venous blood was extracted and 24-hour urine was collected from two groups of patients, turbidimetric immunoassay was used to determine the creatinine (Scr) and cystatin C (CysC) levels in peripheral blood, and rate method was used to determine the β2 microglobulin (β2-MG) and N-acetyl-β-D-glucosaminidase (NAG) levels in urine.

2.4.2. Serum indexes

Before treatment and after 3 months of treatment, 2 mL of fasting peripheral venous blood was extracted from two groups of patients, added in citric acid, let stand at room temperature for the night and centrifuged at 2 500 r/min for 10 min to get supernatant, and the detection indexes are as follows: (1) the peroxidation damage: ELISA method was used to detect reactive oxygen species (ROS), advanced protein oxidation products (AOPPs), catalase (CAT) and total superoxide dismutase (TSOD) levels; (2) inflammatory injury: ELISA method was used to detect pro-inflammatory factors interleukin-8 (IL-8), interleukin-27 (IL-27) and procalcitonin (PCT) levels as well as anti-inflammatory factors interleukin-4 (IL-4), interleukin-10 (IL-10) and interleukin-13 (IL-13) levels.

2.5. Statistical analysis

Data in the study was input in software SPSS20.0, measurement data was in term of mean ± standard deviation (x±s), comparison before and after treatment was by paired t test, comparison between two groups after treatment was by group t test and P<0.05 indicated statistical significance in differences.

3. Results

3.1. Renal function indexes

Comparison of renal function indexes Scr and CysC in peripheral blood as well as renal function indexes β2-MG and NAG levels in urine between two groups of patients is as follows: before treatment, differences in Scr, CysC, β2-MG and NAG levels were not statistically significant between two groups of patients (P>0.05); after 3 months of treatment, Scr, CysC, β2-MG and NAG levels of both groups were lower than those before treatment, and differences within same group were statistically significant (P<0.05); after 3 months of treatment, Scr, CysC, β2-MG and NAG levels of both groups were lower than those of control group, and differences between groups were statistically significant (P<0.05),

<table>
<thead>
<tr>
<th>Groups</th>
<th>Time</th>
<th>Peripheral blood</th>
<th>Urine</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Scr (μmol/L)</td>
<td>CysC (mg/L)</td>
</tr>
<tr>
<td>Observation</td>
<td>Before</td>
<td>126.48±15.19</td>
<td>1.97±0.21</td>
</tr>
<tr>
<td></td>
<td>treatment</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control</td>
<td>After</td>
<td>70.51±7.84a</td>
<td>1.05±0.13</td>
</tr>
<tr>
<td></td>
<td>treatment</td>
<td>125.76±14.32</td>
<td>1.99±0.24</td>
</tr>
<tr>
<td>Observation</td>
<td>After</td>
<td>90.23±9.31a</td>
<td>1.52±0.22</td>
</tr>
<tr>
<td>Control</td>
<td>After</td>
<td>98.72±10.25</td>
<td>1.78±0.23</td>
</tr>
</tbody>
</table>

Compared with same group before treatment, *P<0.05; compared with control group after treatment, **P<0.05.
shown in Table 1.

3.2. Peroxidation damage indexes

Comparison of serum peroxidation damage indexes ROS, AOPPs, CAT and TSOD levels between two groups of patients is as follows: before treatment, differences in serum peroxidation damage indexes ROS, AOPPs, CAT and TSOD levels were not statistically significant between two groups of patients (P > 0.05); after 3 months of treatment, serum ROS and AOPPs levels of both groups were lower than those before treatment while CAT and TSOD levels were higher than those before treatment, and differences within same group were statistically significant (P < 0.05); after 3 months of treatment, serum ROS and AOPPs levels of observation group were lower than those of control group while CAT and TSOD levels were higher than those of control group, and differences between groups were statistically significant (P < 0.05), shown in Table 2.

3.3. Inflammatory injury indexes

Comparison of serum inflammatory injury indexes IL-8, IL-27, PCT, IL-4, IL-10 and IL-13 levels between two groups of patients is as follows: before treatment, differences in serum IL-8, IL-27, PCT, IL-4, IL-10 and IL-13 levels were not statistically significant between two groups of patients (P > 0.05); after 3 months of treatment, serum pro-inflammatory factors IL-8, IL-27 and PCT levels of both groups were lower than those before treatment while anti-inflammatory factors IL-4, IL-10 and IL-13 levels were higher than those before treatment, and differences within same group were statistically significant (P < 0.05); after 3 months of treatment, serum pro-inflammatory factors IL-8, IL-27 and PCT levels of observation group were lower than those of control group while anti-inflammatory factors IL-4, IL-10 and IL-13 levels were higher than those of control group, and differences between groups and differences between groups were statistically significant (P < 0.05), shown in Table 3.

4. Discussion

Diabetic nephropathy is one of the main complications of diabetes mellitus, is currently the second cause of clinical uremia and includes five stages: glomerular hyperfiltration, normal albuminuria stage, early diabetic nephropathy, clinical diabetic nephropathy and end-stage renal failure, there is already urinary albumin in patients with early diabetic nephropathy, but many studies have confirmed that active treatment and intervention in this stage can reverse the illness\[^5\]. Drug therapy is the main treatment for patients with early diabetes. alprostadil, also called prostaglandin E1, has already been successfully applied in cerebrovascular diseases, severe hepatitis, acute pancreatitis, diabetic complications and a variety of other diseases, it has multiple effects such as dilating blood vessels, inhibiting platelet aggregation and reducing blood fat, but the renal protective function of single medication is limited, and other targeted drugs are needed for combined medication\[^6\]. Sorbitol can affect nerve cell function, there is also the accumulation of sorbitol in patients with diabetic nephropathy, and it is an important factor leading to disease progression. Epalrestat is aldose reductase inhibitor, it reversibly inhibits the aldose reductase during glucose conversion to sorbitol to reduce sorbitol generation, and it has played a positive role in the treatment of diabetic nephropathy\[^7\]. In the study, epalrestat and alprostadil form a new combined treatment to protect the patients' renal function from different mechanisms.

There has been a certain degree of abnormal renal function in patients with early diabetic nephropathy, Scr and CysC are the recognized serum indexes to reflect the renal function state, and when renal function continues to decline, the glomerular function to filtrate Scr and CysC declines, and serum Scr and CysC levels increase\[^8\]. β2-MG and NAG levels in urine can also quantitatively express patients’ renal function levels, β2-MG is a kind of small molecular protein that can freely pass through the glomeruli and be re-absorbed by renal tubules, so its urine level is extremely low, the β2-MG re-absorbed by the glomeruli decreases in the case of renal dysfunction, and β2-MG level in urine is consistent with renal function damage extent\[^9\]. NAG is a common index to reflect the renal parenchyma lesion, the NAG level in urine may increase when renal tubular diseases, nephrotic syndrome and diabetic nephropathy occur, and its sensitivity is superior to that of β2-MG\[^10\]. In the
study, the renal function index levels in serum and urine were detected, and it was found that compared with the control group of patients, observation group of patients were with lower Scr and CysC levels in serum as well as β2-MG and NAG levels in urine after 3 months of treatment (P<0.05), confirming that epalrestat combined with alprostadil treatment can effectively optimize the renal function levels.

It is currently believed that oxygen free radicals play an important role in the occurrence and development of early diabetic nephropathy, ROS are massively produced under hyperglycemic state, they cause oxidative damage of renal cellular mitochondrial DNA, block the normal glucose metabolism, and promote glomerular mesangial proliferation, basement membrane thickening and glomerular sclerosis through the reactive oxygen species, cyclooxygenase and other pathways[11,12]. AOPPs are the main products of oxidative metabolism, their levels are consistent with the body’s oxidative damage, and many studies have confirmed that high levels of AOPPs participate in diabetic nephropathy progression[13]. Oxidation/anti-oxidation imbalance is the core mechanism of oxidative damage, the CAT and TSOD, as antioxidant indexes, can inhibit the activity of ROS and neutralize the oxidative effect of AOPPs, and high levels of CAT and TSOD mostly indicate the efficiency and reliability of clinical treatment. In the study, the serum levels of above oxidation and anti-oxidation indexes were detected, and it was found that compared with the control group of patients, observation group of patients were with lower serum ROS and AOPPs levels and higher CAT and TSOD levels after 3 months of treatment (P<0.05), showing that epalrestat combined with alprostadil treatment can inhibit the oxidative stress in patients with early diabetic nephropathy, and this is one of the internal mechanisms of the treatment to protect renal function.

Oxidative stress injury mostly coexists with inflammation injury, and the two interact with each other and generate a vicious circle[14]. The study of Lu et al[15] has confirmed that chronic inflammation is an important pathological change of diabetic nephropathy, and a large number of inflammatory mediators increase the glomerular function damage. The chronic inflammation in patients with early diabetic nephropathy is mainly from imbalance of pro-inflammatory/anti-inflammatory factor expression, both IL-8 and IL-27 are anti-inflammatory factor expression, both IL-8 and IL-27 are detected in serum in the early stage of infectious disease, and it was found that compared with the control group of patients, the renal function index levels in serum and urine were detected, and it was found that compared with the control group, the observation group were with lower serum pro-inflammatory factors IL-8, IL-27 and PCT levels and higher anti-inflammatory factors IL-4, IL-10 and IL-13, as anti-inflammatory factors, are mainly produced by Th2 cells, they can neutralize the excessively produced pro-inflammatory factors and inhibit their recruitment effect on the inflammatory cytokines, the anti-inflammatory factors are massively produced in the early systemic inflammation, and anti-inflammatory factors can be massively consumed because of persistent inflammation and eventually show low levels[17,18]. In the study, the levels of the above pro-inflammatory/anti-inflammatory factors were detected, and it was found that compared with the control group, the observation group were with lower serum pro-inflammatory factors IL-8, IL-27 and PCT levels and higher anti-inflammatory factors IL-4, IL-10 and IL-13 levels (P<0.05), confirming that epalrestat combined with alprostadil can equalize the pro-inflammatory/anti-inflammatory balance in patients with early diabetic nephropathy, and this is another important mechanism for it to protect patients’ renal function.

To sum up, it is concluded as follows: epalrestat combined with alprostadil can protect the renal function and inhibit the peroxidation damage and inflammatory injury in patients with early diabetic nephropathy, and it’s worth popularization and application in clinical practice in the future.

References