The Antiurolithiasic Activity of Aqueous Extract of \textit{Petroselinum Sativum} on Ethylene Glycol-Induced Kidney Calculi in Rats

Saeidi Jafar, Lotfi Mehri, Bozorgi Hadi

\textbf{Abstract}—Forty-eight Wistar rats were divided into 6 groups: Group A as normal control, group B were received 1% ethylene glycol (EG) in drinking water, groups C, D, E and F were received 1% EG in drinking water for 30 days and were also group C treated with 200mg/kg body weight (BW) and group D with 600mg/kg BW of aerial parts, group E with 200mg/kg BW and group F with 600mg/kgBW of root herb extract since the 14th day. The blood samples were collected on days 0 and 30 and serum level of uric acid, urea, calcium and magnesium were evaluated. In day 31, the kidneys prepared for histologic evaluation. The concentrations of serum urea and uric acid were decreased (P<0.05), Serum magnesium concentration was increased and serum calcium concentration was decreased (P<0.05) in treated groups compared with group B. The number of calcium oxalate deposits in group B was higher (P<0.05) than that in group A and in treated groups were decreased (P<0.05) compared with group B. In conclusion \textit{Petroselinum Sativum} has a treatment effect on calcium oxalate stones.

\textbf{Keywords}—\textit{Petroselinum Sativum}, Kidney Stone, Calcium oxalate, Parsley.

\section{I. INTRODUCTION}

Kidney stone formation is a complex process, including supersaturation, nucleation, growth aggregation and retention within the renal tubules [1]. The recurrence of urolithiasis represents a serious problem and thus stone treatment is highly recommended. Additionally, used of ESWL (Extracorporeal shock-wave lithotripsy) method may cause acute renal injury and an increase in stone recurrence [2]. Furthermore, some drugs used to prevent and treatment the disease are not effective in all patients and often have adverse effects that compromise their use in long-term medical treatment. Thus, treatment with herbal drugs has been suggested. The toxic effects of EG have been linked with an increase in free radical production [3]. The \textit{petroselinum sativum} (PS) or parsley, a member of the family of \textit{Umbelliferae}, have been reported to be, antioxidant, anti-inflammatory, antiedema antihypertansive, anti diabetic, antimicrobial and reconstruct kidney tissue after nephrotoxicity [4]. However, there is no evidence for this traditional therapeutic usage. Therefore, we decided to investigate the effect of aqueous extract of parsley on treatment of calcium oxalate calculi in a rat model.

II. MATERIALS AND METHODS

Forty-eight male Wistar rats (200 ± 10 g) were divided randomly into 6 groups: Group A as normal control, group B were received 1% ethylene glycol (EG) in drinking water. Rats of groups C, D, E and F (treated groups) were received 1% EG in drinking water for 30 days. Groups C and D were also treated with 200 and 600mg/kg body weight (BW) of aerial parts and Groups E and F were also treated with 200 and 600mg/kg BW of root aqueous extract of PS since the 14th day through the end of the experiment. The aerial parts and roots of parsley were separated and were dried thoroughly under shad and powdered finely. The powders were suspended in distilled water as aqueous suspension. The extract was dried in an oven with the temperature of 40°C and kept in a refrigerator and was added daily to the drinking water of the rats. The blood samples were collected on days zero and 30 and serum level of uric acid, urea, calcium and magnesium were measured with auto analyzer (BT3000). Data are presented as mean ±SE and were analyzed by one way ANOVA (SPSS for windows, version 16.0). Differences below p<0.05 implied significance. At the end of the experiment (day 31), all rats were killed by guillotine. Then, five micrometer sections of both kidneys were prepared for histological processing. Tubules containing calcium oxalate deposits were counted in 10 light microscopic fields (in $141 \times 10^{-3} \text{ nmm}^2$ of each microscopic fields) [5].

III. RESULTS

Serum magnesium concentration was decreased (P<0.05) and serum calcium concentration, the level of serum urea and uric acid were increased (P<0.05) in group B compared with control group on days 30. The level of serum urea, uric acid and serum concentration of calcium were decreased and serum concentration of magnesium were increased (P<0.05) in treated groups compared with group B on days 30 (table 1, fig. 1, fig. 2). The number of calcium oxalate deposits in 10 microscopic fields in the kidney specimens of group B was significantly higher than that in group A. In treated groups the number of calcium oxalate deposits were decreased (P<0.05) compared with group B. Calcium oxalate crystals in different parts of the renal tubules in the treated groups were smaller in comparison with group B.
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Calcium oxalate crystallization in their kidneys [14] - [15]. These findings indicate that rats supplemented with herb extract were mostly recovered from nephrolithiasis. Serum magnesium was significantly diminished in EG induced urolithic rats. Magnesium complexes with oxalate, thus reducing CaOx super saturation in urine [16].Treatment with aqueous extract of PS reduced the level of serum calcium in rats. This indicates that PS materials mainly glycoside flavonoids act as inhibiting some steps of oxalate synthesis from glycolic acid [17].

Fig.1 The concentration of serum Ca$^{2+}$ and Mg$^{2+}$ (mg/100 cc serum) of control (GgoudA),Ethylene glycol control(Groud B) and treatments groups (C,D,E and F)

Fig.2 The concentration of serum urea and uric acid on day 30 of experiment in control (A and B) and treated groups of rats(C,D,E and F)

Table 1

<table>
<thead>
<tr>
<th>Parameters</th>
<th>Day</th>
<th>Group A</th>
<th>Group B</th>
<th>Group C</th>
<th>Group D</th>
<th>Group E</th>
<th>Group F</th>
</tr>
</thead>
<tbody>
<tr>
<td>Urea (mg/100 cc serum)</td>
<td>0</td>
<td>65.33 ± 4.44</td>
<td>64.33 ± 2.91</td>
<td>71.83 ± 0.95</td>
<td>65.17 ± 0.00</td>
<td>70.33 ± 19.73</td>
<td>66.5 ± 10.86</td>
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<tr>
<td></td>
<td>30</td>
<td>224.25 ± 141.85</td>
<td>141.85 ± 36.97</td>
<td>133.33 ± 13.33</td>
<td>141.85 ± 0.00</td>
<td>114.33 ± 11.43</td>
<td></td>
</tr>
<tr>
<td>Uric acid (mg/100 cc serum)</td>
<td>0</td>
<td>6.85 ± 0.99</td>
<td>6.35 ± 0.5</td>
<td>4.43 ± 0.39</td>
<td>5.58 ± 0.28</td>
<td>5.68 ± 4.1</td>
<td>6.33 ± 2.91</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>4.7 ± 0.28</td>
<td>5.57 ± 0.18</td>
<td>3.05 ± 0.24</td>
<td>4.1 ± 0.34</td>
<td>4.1 ± 0.17</td>
<td>2.87 ± 0.44</td>
</tr>
<tr>
<td>Calcium (mg/100 cc serum)</td>
<td>0</td>
<td>8.95 ± 0.15</td>
<td>8.65 ± 0.21</td>
<td>8.57 ± 0.21</td>
<td>9.33 ± 0.34</td>
<td>9.27 ± 0.24</td>
<td>9.03 ± 0.23</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>8.30 ± 0.15</td>
<td>11.33 ± 0.24</td>
<td>9.62 ± 0.24</td>
<td>10.02 ± 0.24</td>
<td>9.20 ± 0.24</td>
<td>10.19 ± 0.48</td>
</tr>
<tr>
<td>Magnesium (mg/100 cc serum)</td>
<td>0</td>
<td>4.35 ± 0.11</td>
<td>4.33 ± 0.35</td>
<td>4.3 ± 0.18</td>
<td>4.65 ± 0.35</td>
<td>3.67 ± 0.27</td>
<td>4.18 ± 0.27</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>6.01 ± 0.25</td>
<td>3.81 ± 0.18</td>
<td>5.87 ± 0.18</td>
<td>7.05 ± 0.10</td>
<td>4.70 ± 0.27</td>
<td>6.63 ± 0.00</td>
</tr>
<tr>
<td>Number of CaOx crystals (in 10 microscopic fields)</td>
<td>30</td>
<td>16.72 ± 2.22</td>
<td>6.15 ± 1.85</td>
<td>11.95 ± 1.39</td>
<td>10.62 ± 0.45</td>
<td>8.88 ± 0.53</td>
<td></td>
</tr>
</tbody>
</table>

* Indicates a significant difference between treated groups and Group B (P<0.05)

IV. DISCUSSION

Calcium oxalate crystals in urinary tubules can produce damages in the epithelial cells [6]-[7] and consequently, the cells may produce free radicals, inducing heterogenous crystal nucleation and cause aggregation of crystals [8]. Aqueous extract of PS have glycosidea flavonoids such as apiine ,apiol, apigenin , myristine ,tanin, palmitic acid, etc [9] and the flavonoides have antioxidant effects [10].It can be speculated that of the role of the herb extract in treatment of calcium oxalate calculi , is due to the antioxidant effects of the different compounds of the PS [11]. The calcium salts are insoluble at physiological pH inducing calcium oxalate (CaOx) nephrolithiasis[8].We believed that the herb extract provide the optimum pH which it can maintain CaOx particles dispersed in the solution and thus allow them to be eliminated easily from the kidney. The mechanism of herb extract on nephrolithiasis related to increased diuresis via an inhibition of the Na$^-$–K$^+$ pump in renal epithelial cell [12].It probably that the PS extract can disperse CaOx crystals in the solution and eliminate them from the kidney easily. It has been reported that CaOx calculi may have a bacterial origin [13]. Antimicrobial activity of PS materials against natural microflora may be effective in this mechanism. The present results showed that the administration of EG caused statistically increases in the levels of serum calcium and a decrease in the level of serum magnesium. Karadi et al. (2006) and Celik et al. (2007) have shown that magnesium deficiency accelerates renal stone formation in rats and administration of magnesium results in prevention of

The drugs inducing nephrotoxicities are often associated with marked elevations in blood urea and acid uric and acute tubular necrosis .In urolithiasis, the glomerular filtration rate decreases due to the obstruction of the outflow of urine by stones in urinary system. Due to this, the waste products particularly nitrogenous substances such as proteins, urea
and uric acid accumulate in blood [18]. In our study, the nephrotoxicity induced by ethylene glycol, was characterized by elevation of blood urea and uric acid. But these parameters were decreased in the treated groups compared to the group B. The kidney histology revealed a characteristic effect ranging from vascular congestion or haemorrhage to diffuse cortical necrosis. Whereas in all the treated groups, a little and moderate necrosis was also observed in the histopathological sections of the kidneys in the majority of animals, indicating a reduction of the extent of damage done at the tissue level [19]. It suggests that the effect of the extracts could be advantageous in preventing urinary stone retention by reducing renal necrosis and thus inhibit crystal retention. In addition, the aqueous extract of PS was not nephrotoxic at this concentration. Microscopic examination of kidney sections derived from group B (fig. 3) showed polymorphic irregular crystals deposits inside the tubules. Treated groups had increased the normal renal architecture.

![Polymorphic irregular CaOx crystals inside the renal tubules](image)

Our study show that the aqueous extract of PS that contain flavanol glycoside and saponins can prevent or slow down the oxidative damage in epithelial cells of kidney. Further investigation is needed to explore the exact active principles responsible for the antilithiatic activity of aqueous extract of PS and its mechanism of action.

V. ACKNOWLEDGEMENTS

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REFERENCES


