Nephroprotective effect of Corn Silk extract on oxalic acid-induced nephrocalcinosis in rabbit model

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Abstract
Background: Nephrocalcinosis is a state of deposition of calcium phosphate or oxalate in the renal parenchyma. It may occur in patients with renal tubular acidosis, vitamin D intoxication, and hyperparathyroidism. Corn silk was used in traditional Chinese medicine to relieve renal pains. Aim: To evaluate the effect of Corn silk aqueous extract in reducing calcium deposits from renal parenchyma in oxalic acid-induced nephrocalcinosis model.

Methods: Fourteen healthy rabbits were allocated to two groups. Two hours before induction of nephrocalcinosis, one group received water and the other received aqueous extract of corn silk and continued feeding for ten days. Blood samples were collected for biochemical analysis before induction and in the fifth and tenth post-induction day. Urine samples were taken to estimate urinary Ca+2 levels and crystals. The histopathological examination was carried to check for crystal deposits in renal tissues.

Results: Corn silk aqueous extract produced a significant reduction of blood urea nitrogen(5.2±0.08 vs 7.3±0.2) mmol/l, serum creatinine (85.9±0.2 vs 97.3±0.5) mmol/l and serum Na+ levels (137±0.2 vs 142.16±0.7) mmol/l with non-significant reduction in serum K+ (4.0±0.02 vs 4.2±0.05). There is a significant reduction in calcium deposition in renal parenchyma in comparison to the control group after ten days of treatment.

Conclusion: Corn silk had a significant diuretic effect that accelerates the excretion of urinary calcium.

INTRODUCTION
Nephrocalcinosis (NC) is a state of deposition of calcium in the form of phosphate or oxalate in the renal parenchyma that can impair kidney function [1]. It is liable to occur in patients with renal tubular acidosis, hyperparathyroidism, vitamin D intoxication and healing of renal tuberculosis [2] The fragments of calcium salts may break freely from the kidney to provide nuclei for the formation of different types and sizes of stones [3].

In traditional Chinese medicine, some medicinal plants like corn silk, barley and celery were used to relieve renal pains [4]. Corn silk resembles soft threads of either light green or yellow-brown in color. It contains [5]: proteins, carbohydrates, volatile oils, steroids such as sitosterol, saponins, and flavonoids. Extract of corn silk showed an anti-oxidative [6] and anti-TNF activity [7].

Few studies were made determining the diuretic effect of Barley [8] and Celery [9], but without available data to evaluate the possible nephroprotective effect of corn silk. Therefore, this study was made to evaluate this potential effect in an experimental model of Nephrocalcinosis.
MATERIALS AND METHODS

Fourteen local domestic healthy rabbits weighing 900 to 1200 grams were used in this study, which was approved by Animal Ethics Committee of the college of pharmacy, Al-Yarmouk University (Approval No.AEC/31/10/CPAYU). The rabbits were supplied by the animals' house of college of medicine. They were housed in separated cages, which were provided with a wide wire mesh floor at a controlled temperature of 27±2°C with a 12-hour light/dark cycle. They were fed standard oxoid pellets and water ad libitum.

The animals were allocated to two groups (seven animals in each) and were given the following as a single daily dose for 10 days (at 9 a.m.);

**G1** (control group) - received 3 ml/kg of distilled water

**G2** - received 1gm/kg of aqueous extract of Corn silk [10]

At 9 a.m. of 10th day, the last doses were given and the rabbits were fasted for 24 hours.

At 11 a.m. of next day, all animals were given 333 mg/kg of oxalic acid (H&W England) as a single dose per gastric tube for induction of NC [11]

Blood samples were collected from a marginal ear vein for biochemical analysis and renal functions, before induction of NC to determine the normal values of blood urea nitrogen (BUN), serum creatinine, Na⁺ and K⁺ using spectrophotometer and in the 1st, 5th and 10th day after induction. Urine samples were taken from the animals by catheterization after anesthetizing them in the last day of the study to determine the urinary Ca²⁺ levels and present crystals. The histopathological examination was carried to check for crystal deposits in renal tissues by using a polarized microscope after fixation and staining the specimens [12] The obtained results were collected for analysis and assessment. Significance was set at P<0.05.

RESULTS

The results of this study revealed significant elevation in the levels of BUN, both serum creatinine and K⁺ with significant reduction of serum Na⁺ levels in the control group as compared to the levels of pre-induction state [Table 1].

<table>
<thead>
<tr>
<th>Analyte</th>
<th>Measured levels (mmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td>BUN</td>
<td>4.0±0.07</td>
</tr>
<tr>
<td>S. creatinine</td>
<td>65.0±8.9</td>
</tr>
<tr>
<td>S. K⁺</td>
<td>3.3±0.8</td>
</tr>
<tr>
<td>S. Na⁺</td>
<td>160.0±4.0</td>
</tr>
<tr>
<td>Urine Ca²⁺</td>
<td>2.05±0.07</td>
</tr>
</tbody>
</table>

The results of Zea Mays extract (group 2) showed significant reduction of BUN levels (5.2 ± 0.08 vs. 7.3 ± 0.2) mmol/L, serum creatinine (85.9 ± 0.2 vs. 97.3 ± 0.5) mmol/L with P<0.05, and insignificant reduction in serum K⁺ (4.0±0.02 vs 4.2±0.05) mmol/l with significant decrease in serum Na⁺ levels (137 ± 0.2 vs. 142.16 ± 0.7) mmol/L with P< 0.05 in comparison to the control group after one day and ten days when results became more evident [Table 2-3].

<table>
<thead>
<tr>
<th>Group</th>
<th>BUN (mmol/L)</th>
<th>Creatinine (mmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>After 1 day</td>
<td>After 5 day</td>
</tr>
<tr>
<td>Oxalic acid</td>
<td>7.3±0.2</td>
<td>7.9±1.1</td>
</tr>
<tr>
<td>Corn silk</td>
<td>5.2±0.08</td>
<td>5.1±0.3</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Group</th>
<th>Serum K⁺ (mmol/L)</th>
<th>Serum Na⁺ (mmol/L)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>After 1 day</td>
<td>After 5 day</td>
</tr>
<tr>
<td>Oxalic acid</td>
<td>4.2±0.055</td>
<td>1±0.24</td>
</tr>
<tr>
<td>Corn silk</td>
<td>4.0±0.023</td>
<td>9±0.1</td>
</tr>
</tbody>
</table>
Table 4. Mean urine Ca\(^{2+}\) levels of the studied groups measured 10 days after induction of NC

<table>
<thead>
<tr>
<th>Group</th>
<th>Urine Ca(^{2+}) levels (mmol/L)</th>
<th>Crystals in urine examination</th>
</tr>
</thead>
<tbody>
<tr>
<td>- (negative control)</td>
<td>2.05±0.07</td>
<td>few amorphous urate</td>
</tr>
<tr>
<td>1 (Oxalic acid)</td>
<td>4.11±0.03</td>
<td>amorphous urate, uric ac stellar phosphate, calcium oxalate id</td>
</tr>
<tr>
<td>2 (Oxalic acid + Corn silk)</td>
<td>5.06±0.08</td>
<td></td>
</tr>
</tbody>
</table>

Examination of urine samples at the end of the experiment (10 days) showed that there was a significant elevating effect of corn silk in urinary Ca\(^{2+}\) excretion than normal values [12] and a significant reduction of its concentration in renal parenchyma after histological examination of renal tissue samples [Table 4] [Figures 1-2].

**DISCUSSION**

Oxalic acid (a highly oxidizing and strong Ca\(^{2+}\) chelator) was used for induction of NC using a single large dose (333mg/kg) [11] In this model of NC, there was a significant elevation in BUN and serum creatinine levels observed after induction of Nephrocalcinosis [Tables 2 and 3]. The idea to use some herals like corn silk is to evaluate its effect in improvement of NC. Corn silk contains many active ingredients like flavonoids, volatile oil, saponins, and allantoin besides some minerals like calcium, potassium, magnesium and sodium. It produced a significant lowering effect in BUN and serum creatinine levels with an increase of urinary Ca\(^{2+}\) levels than normal values [12] and a reduction of its concentration in renal tissue [Figures 1-3]. Because of its high concentration of potassium, it becomes a powerful diuretic, which encourages the body to flush out toxins by increasing urination and Ca\(^{2+}\) excretion [13] Unlike other diuretics, the high level of potassium offsets the potassium loss normally caused with increased urination when in use [14]

**CONCLUSION**

Zea mays has a significant diuretic and attenuating effect in reducing calcium deposits from renal tissues.

**REFERENCES**


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