Acid-Base Status Determines Cyclosporine-Induced Hypercalciuria

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ABSTRACT

Objective. Cyclosporine (CsA) causes tubular dysfunction characterized by polyuria, calcium wasting, distal tubular acidosis, and hyperkalemia. The hypercalciuria induced by CsA administration is associated with an inhibition of calbindin D$_{28k}$ expression. It has also been shown that chronic metabolic alkalosis increased the expression of Ca$^{2+}$/H$^{+}$ transport proteins accompanied by diminished urine Ca$^{2+}$ excretion. The aim of this study, therefore, was to determine the effect of acid-base status on CsA-induced hypercalciuria.

Methods. Experiments were performed on male Sprague-Dawley rats. Metabolic alkalosis and acidosis were induced respectively by adding 0.28 mol/L NaHCO$_3$ and 0.28 mol/L NH$_4$Cl in the drinking water for 7 days; control rats received regular tap water. Seven days after NaHCO$_3$ or NH$_4$Cl administration, rats were treated with CsA (25 mg/kg, IP) daily for 14 days. To estimate glomerular filtration rate (GFR) over time, animals were placed in metabolic cages. Fractional urinary calcium excretion was determined by standard formula.

Results. The CsA group showed decreased serum calcium and increased fractional urinary calcium excretion compared with the control group. Creatinine clearance was also significantly reduced. Metabolic alkalosis alone did not affect GFR, but significantly prevented an increase in fractional urinary calcium excretion induced by CsA, whereas chronic metabolic acidosis resulted in the exact opposite effect.

Conclusions. It is essential for nephrologists to fully understand the mechanisms of CsA-induced renal injury. In this study, metabolic alkalosis reduced CsA-induced hypercalciuria. Further studies are needed to elucidate whether this effect may be achieved pharmacologically by the expression of Ca$^{2+}$/H$^{+}$ transport proteins.

Cyclosporine (CsA) is shown to improve patient and graft survival rates. However, the clinical use of CsA is often limited by nephrotoxicity, which remains a major problem. CsA causes tubular dysfunction characterized by polyuria, calcium wasting, distal tubular acidosis, and hyperkalemia. The hypercalciuria induced by CsA administration is associated with dysregulation of parathyroid hormone. The Ca$^{2+}$ binding protein so-called calbindin plays a significant role in the process of Ca$^{2+}$ transport. Calbindin D$_{28k}$ is expressed in the distal nephron segments of the rat kidney and is assumed to take part in the process of Ca$^{2+}$ reabsorption. Yang et al$^3$ and Steiner et al$^4$ have shown that CsA treatment could inhibit calbindin D$_{28k}$ which was accompanied by a significant decrease in serum Ca$^{2+}$ concentration and an increase in urinary Ca$^{2+}$ excretion. This suggests that CsA-mediated suppression of calbindin D$_{28k}$ production may be a critical factor in renal Ca$^{2+}$ wasting.$^1$ It has also been shown that chronic metabolic acidosis results in renal Ca$^{2+}$ wasting, whereas chronic metabolic alkalosis is known to exert the reverse effect. Nijenhuis et al$^5$ showed that these adaptations are mediated at least in part by the renal Ca$^{2+}$ transport proteins and that chronic metabolic alkalosis increased the expression of Ca$^{2+}$ transport proteins accompanied by diminished urine Ca$^{2+}$ excretion. The aim of this study,
Seven days after NaHCO3 or NH4Cl administration, rats were drinking water for 7 days; control rats received regular tap water. With the control group. Serum [HCO3-] was not significantly different compared with the other groups.

**DISCUSSION**

It is essential for nephrologists to fully understand the mechanisms of CsA-induced renal injury as pharmacological intervention may delay the progression of chronic CsA nephrotoxicity. In this study, metabolic alkalosis reduced CsA-induced hypercalciuria. Previous studies showed that higher doses of CsA are required in rats than in humans to cause renal damage. This effect was probably due to the lower sensitivity of rats to CsA. Down-regulation of renal calbindin D28K has been proposed to be the pathogenetic mechanism for CsA-induced hypercalciuria in rats. Nijenhuis et al. showed that chronic metabolic alkalosis that was induced by NaHCO3 administration for 6 days increased the expression of Ca2+ transport proteins and was accompanied by diminished urinary Ca2+ excretion. In our studies, metabolic alkalosis induction which was demonstrated by an elevated plasma pH and bicarbonate reduced CsA-induced hypercalciuria. Prevention of renal calcium excretion in alkalotic rats may be mediated at least in part by mechanism(s) dependent on calbindin D28K. Another potential mechanism is that CsA-induced renal tubular acidosis may enhance hypercalciuria, as pH is an important regulator of distal calcium reabsorption. In conclusion, these data suggest that acid-base status may be a critical factor in renal Ca2+ wasting by CsA. These results have important clinical implications in the management of patients who receive calcineurin inhibitors. Further studies are needed to elucidate whether this effect may be achieved pharmacologically by the expression of Ca2+ transport proteins.

**REFERENCES**


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**Table 1. Data on Renal Function, Urinary Calcium Excretion, and Acid-Base Status in 6 Groups of Rats**

<table>
<thead>
<tr>
<th>Group</th>
<th>GFR (mL/min/100 g)</th>
<th>FE% Calcium</th>
<th>HCO3- (mmol/L)</th>
<th>pH</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>0.84 ± 0.015†</td>
<td>0.14 ± 0.004†</td>
<td>23.97 ± 0.77†</td>
<td>7.4 ± 0.01†</td>
</tr>
<tr>
<td>CsA</td>
<td>0.67 ± 0.037*</td>
<td>1.49 ± 0.22*</td>
<td>19.09 ± 0.49*</td>
<td>7.39 ± 0.013</td>
</tr>
<tr>
<td>NaHCO3</td>
<td>0.91 ± 0.047†</td>
<td>0.13 ± 0.005†</td>
<td>26.6 ± 0.49†</td>
<td>7.46 ± 0.030*</td>
</tr>
<tr>
<td>NH4Cl</td>
<td>0.81 ± 0.010†</td>
<td>1.24 ± 0.07†</td>
<td>16.32 ± 0.53†</td>
<td>7.35 ± 0.014</td>
</tr>
<tr>
<td>CsA + NaHCO3</td>
<td>0.80 ± 0.031†</td>
<td>0.51 ± 0.10†</td>
<td>26.89 ± 0.44†</td>
<td>7.45 ± 0.016*</td>
</tr>
<tr>
<td>CsA + NH4Cl</td>
<td>0.64 ± 0.037†</td>
<td>1.83 ± 0.05†</td>
<td>15.37 ± 0.60†</td>
<td>7.32 ± 0.023†</td>
</tr>
</tbody>
</table>

Data are presented as means ± SEM (n = 5 in each group). *P < .05 compared with control. †P < .05 compared with CsA group.