Renal Transport of Lysine and Arginine in Cystinuria

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KATO, T. Renal Transport of Lysine and Arginine in Cystinuria. Tohoku J. exp. Med., 1983, 139 (1), 9-16 — To study the defective transport mechanism of cystinuria, renal tubular reabsorption of lysine or arginine in normal and cystinuria subjects was investigated by increasing the filtered load employing intravenous amino acid infusion. In the normal group the amino acid reabsorption rose with increases of the filtered load and reached a maximum (Tm). In the cystinuria group the elevation of amino acid reabsorption was poor at low filtered loads and some of the reabsorption rates fell below zero, whereas the tubular transport proceeded at a normal rate with a great increase of the filtered load. This might be explained as follows: At low filtered loads the filtered amino acid in the tubular lumen in patients with cystinuria is not absorbed into the cell because of a transport defect of the luminal membrane of the tubular cell, causing a large amount of urinary amino acid excretion. At high filtered loads the accumulated intraluminal amino acid permeates the tubular cell by a passive diffusion and is transported to the capillary across the intact basolateral membrane, which in turn only brings about a small urinary loss of amino acid. The infusion of lysine or arginine depressed the percentage of tubular reabsorption of other dibasic amino acids in both groups. In the cystinuria group the percentage of the dibasic amino acid reabsorption dropped sharply with an initial load of the inhibitor, but no more depression of the percentage of reabsorption occurred with further loads of the inhibitor.

Cystinuria is a hereditary disorder characterized by abnormalities in the transport of dibasic amino acids lysine, arginine and ornithine, and cystine in both the intestinal tract and renal tubule (Scriver and Rosenberg 1973). Subjects with this disease excrete large amounts of the dibasic amino acids and cystine in the urine and often form cystine calculi in the urinary tract (Crawhall and Watts 1968). Dent and Rose (1951) postulated that the dibasic amino acids and cystine have a common transport system which is defective in cystinuria. Lysine or arginine infusion in man (Robson and Rose 1957; Kato 1977) increases the urinary excretion of other dibasic amino acids and cystine, supporting Dent and Rose’s hypothesis. However, in vitro studies with renal cortical slices (Rosenberg et al. 1962; Fox et al. 1964) have shown that the dibasic amino acids have a common transport system that is not shared by cystine. In addition, hyperexcretion of the dibasic amino acids without hypercystinuria is seen in lysinuric protein intolerance (LPI), a rare inherited disorder with a transport defect of the
dibasic amino acids only in both the gut and kidney (Simell et al. 1975). Recently, Segal et al. (1977) and Foreman et al. (1980), using isolated rat renal tubules, have demonstrated two transport systems for cystine. The low Km system, which predominates at low substrate concentrations, interacts with the dibasic amino acids, while the high Km system predominating at high concentrations does not, suggesting that cystine not only shares a common transport system with the dibasic amino acids but that it also has a separate transport system from the dibasic amino acids. In vitro experiments with kidney slices (Rosenberg et al. 1967) and skin fibroblasts (Groth and Rosenberg 1972) have demonstrated two different transport processes of the dibasic amino acids, one of which predominates at low substrate concentrations and the other at high substrate concentrations. The low-concentration system of the dibasic amino acids would correspond to the low Km system of cystine, and the high-concentration system would separate from the high Km system of cystine. Kato (1977), studying in vivo renal clearance of patients with cystinuria, has postulated a transport defect of the low-concentration system of the dibasic amino acids in cystinuria.

Coicadan et al. (1980) have suggested a transport defect of lysine in the luminal membrane of the intestinal brush border cell in cystinuria, and Desjeux et al. (1980) and Rajantie et al. (1980, 1981) have demonstrated a transport defect of the dibasic amino acids in the basolateral membrane of both the gut mucosa and renal tubule in LPI, indicating the location of the defective transport site by the cell membrane. In the present experiments, in vivo renal transport of lysine and arginine in patients with cystinuria was examined to study the defective transport mechanism of the tubular cell of this disease.

**Subjects and Methods**

Five control subjects and six cystinuria patients with a history of cystine stone formation in the urinary tract were studied. Urinary excretion of the dibasic amino acids and cystine in each subject is shown in Table 1. Informed consent to the study was obtained from their parents.

All experiments were performed early in the morning under fasting conditions. Sufficient water was given orally to cause a diuresis exceeding 8 ml per min. Each clearance study was done for 20 min where urine was collected by spontaneous voiding and blood was drawn at the midpoint of the period. Endogenous creatinine clearance was used to estimate the glomerular filtration rate. After a control clearance test, L-lysine monohydrochloride or L-arginine monohydrochloride was infused as a 1 M solution or its dilution into a peripheral vein within 5 min followed by a sustaining infusion. The priming doses of amino acid were 0.7 to 1.5 mmoles per kg of body weight, and the sustaining doses 1 to 10 μmoles per kg of body weight per min. Three or four consecutive or intermittent clearance tests were performed during the sustaining infusion.

Amino acid in urine and serum was estimated by ion-exchange chromatography (Spackman et al. 1958) with a Hitachi amino acid analyzer. Creatinine was measured by the method of Bonsnes and Taussky (1945). Urinary excretion, renal clearance and tubular reabsorption of amino acid were calculated according to Scriver and Davies (1965).
TABLE 1. Urinary excretion of dibasic amino acids and cystine in normal and cystinuric subjects

<table>
<thead>
<tr>
<th>Subject</th>
<th>Age</th>
<th>Sex</th>
<th>Lysine</th>
<th>Arginine</th>
<th>Ornithine</th>
<th>Cystine</th>
</tr>
</thead>
<tbody>
<tr>
<td>N1</td>
<td>14</td>
<td>M</td>
<td>15.5</td>
<td>3.64</td>
<td>5.88</td>
<td>10.9</td>
</tr>
<tr>
<td>N2</td>
<td>15</td>
<td>F</td>
<td>80.3</td>
<td>6.98</td>
<td>3.07</td>
<td>18.1</td>
</tr>
<tr>
<td>N3</td>
<td>9</td>
<td>F</td>
<td>19.3</td>
<td>8.71</td>
<td>6.03</td>
<td>17.3</td>
</tr>
<tr>
<td>N4</td>
<td>11</td>
<td>M</td>
<td>17.9</td>
<td>6.68</td>
<td>2.60</td>
<td>7.75</td>
</tr>
<tr>
<td>N5</td>
<td>14</td>
<td>M</td>
<td>23.2</td>
<td>8.50</td>
<td>5.50</td>
<td>10.2</td>
</tr>
<tr>
<td>C1</td>
<td>15</td>
<td>M</td>
<td>1,002</td>
<td>1,065</td>
<td>319</td>
<td>670</td>
</tr>
<tr>
<td>C2</td>
<td>9</td>
<td>F</td>
<td>2,289</td>
<td>1,526</td>
<td>493</td>
<td>379</td>
</tr>
<tr>
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<td>10</td>
<td>M</td>
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<td>1,164</td>
<td>598</td>
<td>323</td>
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<tr>
<td>C4</td>
<td>19</td>
<td>M</td>
<td>837</td>
<td>540</td>
<td>137</td>
<td>347</td>
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<tr>
<td>C5</td>
<td>15</td>
<td>F</td>
<td>1,213</td>
<td>1,058</td>
<td>431</td>
<td>488</td>
</tr>
<tr>
<td>C6</td>
<td>15</td>
<td>M</td>
<td>1,353</td>
<td>502</td>
<td>205</td>
<td>270</td>
</tr>
</tbody>
</table>

N1–5, normal subjects; C1–6, cystinuric subjects

RESULTS

The effect of increased filtered load on tubular reabsorption is shown in Fig. 1. When the filtered load of lysine or arginine was increased by intravenous infusion, the tubular reabsorption of amino acid in the control group rose with increases of the filtered load and reached a maximum (Tm). On the other hand, in the cystinuria group the reabsorption of lysine was not raised as much as that of the...
controls at low filtered loads and the reabsorption rate sometimes gave negative values showing a tubular secretion of amino acid. The elevation of arginine reabsorption was also very poor under these conditions. However, when the filtered load was fully increased the reabsorption rate of each amino acid of the patients reached the level of the controls. This suggests that in the patients with cystinuria the renal transport of the dibasic amino acids was defective at low filtered loads, while the tubular reabsorption proceeded at a normal rate at high filtered loads.

The inhibitory effects of lysine or arginine on the percentage of other dibasic amino acid reabsorption are shown in Figs. 2 and 3. The percentage of reabsorption

Fig. 2. Percentage of tubular reabsorption of arginine (A) and ornithine (B) plotted against filtered lysine in control (○) and cystinuric subjects (●). The presumed depression curve of controls was drawn using the model made by Scriver (1968).

Fig. 3. Percentage of tubular reabsorption of lysine (A) and ornithine (B) plotted against filtered arginine in control (○) and cystinuric subjects (●).
of depressed amino acid in the control subjects gradually decreased with increases of the inhibitor load, leaving about 50 to 70% of the reabsorption capacity uninhibited. In the cystinuria patients the percentage of the tubular reabsorption of depressed amino acid also dropped but sharply with the initial load of the inhibitor, but no more depression of amino acid reabsorption was found with further loads of the inhibitor.

**DISCUSSION**

Information about in vivo renal handling of amino acid has been obtained by studying the clearance of amino acid at both endogenous plasma concentrations and elevated amino acid levels employing intravenous infusion. Since Pitts (1943) first reported the titration curve of glycine in dog, renal transport of amino acid has been studied in animals and man using the in vivo clearance method (Young and Freedman 1971). The slope of the reabsorption curve of lysine and arginine in the present normal subjects was nearly the same as that observed in animals (Wright et al. 1947; Boorman 1971), showing a little wide splay and Tm that is often seen in other amino acid titration curves (Young and Freedman 1971). The renal transport mechanism of human kidney may be similar to that of animals.

Desjeux et al. (1980) and Rajantie et al. (1980, 1981) have demonstrated that the defective transport site of the dibasic amino acids in lysinuric protein intolerance (LPI) is located in the basolateral membrane of both the intestinal and renal tubular cells, and Coicadan et al. (1980) have postulated a transport defect of lysine in the luminal membrane of the gut mucosa in cystinuria. It is likely that the defective transport site of the dibasic amino acids in cystinuria is also located in the luminal membrane of the tubular cell.

In the present study the tubular reabsorption of lysine and arginine in the cystinuria patients did not rise as much as that of the control subjects and some of the reabsorption rates gave negative values at low filtered loads, whereas the tubular transport of each amino acid was done at a normal rate with a great increase of the filtered load, showing nearly the same finding as that obtained in the earlier experiments (Kato 1977). An explanation for this might be proposed. When the filtered load is low, the filtered amino acid in the tubular lumen in patients with cystinuria is not absorbed into the tubular cell because of a transport defect of the luminal membrane and directly excreted into the urine, producing a large amino acid excretion in the urine. However, when the filtered load is fully increased the extremely accumulated intraluminal amino acid permeates the cell by a passive diffusion, which is rapidly transported to the capillary across the intact basolateral membrane, which in turn only brings about a small urinary leakage of amino acid under this condition.

It is interesting to compare the present result with that observed in LPI in which the transport defect of the dibasic amino acids is assumed to exist in the basolateral membrane of the tubular cell. When the tubular reabsorption of dibasic amino acid of a patient with LPI was raised by increasing the filtered load, in
contrast to the present result, the amino acid reabsorption ability was present at low filtered loads, while with a marked increase of the filtered load the amino acid reabsorption of the patient was grossly impaired (Kato et al. 1982). A suitable explanation for this was presented as follows: The filtered amino acid in the lumen in LPI is accumulated into the cell across the intact luminal membrane at low filtered loads, leading to a small amino acid excretion in the urine, while at high filtered loads the saturated intracellular amino acid, which is not transported to the capillary because of a basolateral transport defect, leaks back to the lumen, causing a large urinary excretion of amino acid. At physiological conditions the renal transport defect of the dibasic amino acids in cystinuria seems more severe than in LPI because endogenous renal clearance of the dibasic amino acids in cystinuria increases more (Morin et al. 1971; Simell et al. 1975). This might be attributable to the differences of the defective transport mechanisms between the two diseases as mentioned above.

As the dibasic amino acids share a common transport system, this system may be saturated by one of these amino acids, that brings about an inhibition of the renal transport of others (Webber et al. 1961). In the present inhibition study the administration of lysine or arginine depressed the percentage of tubular reabsorption of other dibasic amino acids in both groups. The percentage of amino acid reabsorption of the cystinuria group, which already grossly decreased before the inhibitor load, further dropped sharply with the initial load of the inhibitor. But the percentage of the reabsorption remained fixed at the initial level although the inhibitor load was increased. This would be explained as follows: The filtered inhibitor in the lumen occupies the remaining intact transport site of the luminal membrane, depressing the absorption of other intraluminal dibasic amino acids. But, as this intact transport site is very small as compared to that of controls it is rapidly saturated by a small amount of the inhibitor, that causes no more depression of the tubular reabsorption with further loads of the inhibitor.

The present study might propose a hypothesis for the renal tubular transport of the dibasic amino acid. The transport system of the dibasic amino acids located in the luminal membrane of the tubular cell, which is shared with cystine, is defective in cystinuria, and the transport system in the basolateral membrane, which separates from cystine, is defective in LPI. The defect of amino acid transport of the luminal membrane seemingly produces a remarkable impairment when the filtered load is at low levels, and the defect of the basolateral membrane seemingly produces a severe disorder at high amino acid loads.

References


renal clearance of essential amino acids: Arginine, histidine, lysine and methionine. 

_ Amer. J. Physiol., 149_, 130–134.