Emerging role of Intercalated cells in renal salt handling

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Final regulation of acid-base excretion occurs in the CCD

Blood

NaCl absorption
K⁺ secretion

Acid secretion

Base secretion

Urine

\( V_{te} \)

\( \text{Principal cell} \)

\( \text{ENaC} \)
\( \text{Na}^+ \)

\( \text{ROMK} \)
\( \text{K}^+ \)

\( \text{Cl}^- \)

\( \text{\( \beta\)-Intercalated cell} \)

\( \text{AE1} \)
\( \text{HCO}_3^- \)

\( \text{\( \alpha\)-Intercalated cell} \)

\( \text{ATP} \)
\( \text{Na}^+ \)

\( \text{Pds} \)
\( \text{Cl}^- \)

\( \text{\( H^+ \)} \)
Disruption of \( Pds \) blocks \( \text{Cl}^- \) absorption in the CCD

Wall et al. Hypertension 2004
This indicates that Cl\(^-\) absorption is transcellular and occurs through \(\beta\)-IC
*P*ds disruption protects against mineralocorticoid-induced hypertension

Verlander et al. Hypertension 2003
CCD microperfused *in vitro*
Beyond ENaC, the CCD exhibit a NCC-like transporter

Leviel et al. JCI 2010 in press
And HCTZ increases urinary Na excretion in NCC\(^{++/+}\) ... but also in NCC\(^{-/-}\) mice.
Thiazide-sensitive absorption is bicarbonate dependent

Leviel et al. JCI 2010 in press
Thiazide-sensitive Na+ transport is bicarbonate and Cl⁻-dependent

Leviel et al. JCI 2010 in press
SLC4 superfamily

In: $\text{Na}^+$, 2$\text{HCO}_3^-$ 
Out: $\text{Na}^+$, $3\text{HCO}_3^-$, $\text{Cl}^-$ 

Diagram showing the transport of ions in and out of the cell. The diagram includes symbols for $\text{Na}^+$, $\text{Cl}^-$, and $\text{HCO}_3^-$.
Ndce disruption abolishes thiazide-sensitive Na transport

Amiloride-resistant $J_{NaCl}$

Amiloride-resistant $J_{NaCl}$
Two Na\(^+\) transporting systems coexist in mouse CCD

**ENaC-mediated “Na\(^+\)/K\(^+\) exchange**
- Amiloride-sensitive
- HCTZ-resistant
- Electrogenic
- Located in PC

**Pds/NDCBE-mediated NaCl reabsorption:**
- HCTZ-sensitive
- Amiloride-resistant
- Electroneutral
- Located in IC
dRTA a model of global IC dysfunction
Renal Potassium Wasting in Renal Tubular Acidosis (RTA)

ITS OCCURRENCE IN TYPES 1 AND 2 RTA DESPITE SUSTAINED CORRECTION OF SYSTEMIC ACIDOSES

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From the Departments of Medicine and Pediatrics, University of California, San Francisco, California 94122

**Figure 3** Effect of progressively reducing potassium supplements on serum potassium concentration and urinary potassium excretion in a patient with classic renal tubular acidosis (L. C. S.) in whom correction of acidosis was sustained.
Impaired Renal Conservation of Sodium and Chloride during Sustained Correction of Systemic Acidosis in Patients with Type 1, Classic Renal Tubular Acidosis

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FIGURE 2 Relationships between serum sodium concentration, body weight, and the dietary intake and urinary excretion of sodium before and after restricting sodium intake in two patients (A. R., left; C. V., right) with classic RTA in whom correction of systemic acidosis was sustained with potassium bicarbonate therapy.

Conclusion:

dRTA is a tubulopathy leading to tubulo-interstitial nephropathy because of nephrocalcinosis
Renal adaptation to NaCl restriction was delayed.
Mice developed a marked volume contraction

![Graph showing urinary aldosterone levels over time](image)

- **Atp6v1b1**$^{+/+}$
- **Atp6v1b1$^{-/-}$**

![Bar graph showing plasma renin concentration](image)

- WT: 0.3% Na$^+$
- KO: 0.3% Na$^+$
- WT: 0% Na$^+$
- KO: 0% Na$^+$

![Bar graph showing proteins](image)

- WT: 0.3% Na$^+$
- KO: 0.3% Na$^+$
- WT: 0% Na$^+$
- KO: 0% Na$^+$

*p-values:*
- Plasma renin: p<0.001, p=0.02, p=0.004
- Proteins: p=0.005, p=0.005
Mice exhibited renal loss of K⁺, which was majored by Na⁺ restriction leading to hypokalemia.
Proton pump disruption also leads to a urinary concentrating defect.

[Graphs showing changes in urinary output and osmolality over time with different Na+ concentrations and genotypes.]
We expected that H+-pump disruption blocked the function of pendrin: Is it the case?
Pds disruption impairs aldosterone-mediated regulation of ENaC

In pendrin null mice:

Total γ ENaC abundance is reduced ~50%

70 kD γ ENaC abundance is reduced ~70%

From Kim et al Am J Physiol Renal Physiol. 2007
Despite secondary hyperaldosteronism, we observed dysregulation of ENaC in Atp6v1b1⁻/⁻ mice similar to that of Pds KO mice.
Principal cells also exhibited a marked decrease in AQP2 (which can explain polyuria)
Contrasting with CCDs, OMCD a segment devoid of Pendrin exhibited normal response to aldosterone.
PGE2, a factor known to block Na+ and water transport in the renal tubule was elevated.
Blockade of PGE2 production did improve the expression of most proteins except Pendrin

### Cortex

<table>
<thead>
<tr>
<th></th>
<th>Basal state</th>
<th></th>
<th>48h indomethacin</th>
<th></th>
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</thead>
<tbody>
<tr>
<td></td>
<td>WT (n=7)</td>
<td>KO (n=7)</td>
<td>p value</td>
<td>WT (n=6)</td>
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<tr>
<td><strong>Pds</strong></td>
<td>100 ± 11.8</td>
<td>13.3 ± 3.3</td>
<td>&lt;0.0001</td>
<td>100 ± 3.3</td>
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<tr>
<td><strong>ENaC</strong></td>
<td>100 ± 15.7</td>
<td>36.8 ± 3.0</td>
<td>0.0019</td>
<td>100 ± 8.5</td>
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<tr>
<td><strong>γENaC 85kD</strong></td>
<td>100 ± 9</td>
<td>58.7 ± 8.1</td>
<td>0.0055</td>
<td>100 ± 9.7</td>
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<tr>
<td><strong>γENaC 70kD</strong></td>
<td>100 ± 8.3</td>
<td>41.2 ± 7.2</td>
<td>0.0008</td>
<td>100 ± 11.2</td>
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<tr>
<td><strong>BK</strong></td>
<td>100 ± 9</td>
<td>163.3 ± 5.2</td>
<td>&lt;0.0001</td>
<td>100 ± 3.5</td>
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<tr>
<td><strong>AQP2 37kD</strong></td>
<td>100 ± 4.0</td>
<td>85.0 ± 3.5</td>
<td>0.015</td>
<td>100 ± 23.5</td>
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<tr>
<td><strong>AQP2 25kD</strong></td>
<td>100 ± 7.2</td>
<td>191.7 ± 5.8</td>
<td>&lt;0.0001</td>
<td>100 ± 7.6</td>
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### Outer medulla

<table>
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<tbody>
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<td>WT (n=7)</td>
<td>KO (n=7)</td>
<td>p value</td>
<td>WT (n=6)</td>
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<tr>
<td><strong>ENaC</strong></td>
<td>100 ± 18.9</td>
<td>231.6 ± 13.1</td>
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<td>100 ± 28.2</td>
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<td><strong>ENaC 85kD</strong></td>
<td>100 ± 12</td>
<td>148 ± 10</td>
<td>0.012</td>
<td>100 ± 18.4</td>
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<tr>
<td><strong>ENaC 70kD</strong></td>
<td>100 ± 19</td>
<td>407 ± 53</td>
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<td>100 ± 14.1</td>
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<tr>
<td><strong>BK</strong></td>
<td>100 ± 10</td>
<td>258.8 ± 14</td>
<td>&lt;0.0001</td>
<td>100 ± 2.9</td>
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<tr>
<td><strong>AQP2 37kD</strong></td>
<td>100 ± 7</td>
<td>22 ± 7</td>
<td>&lt;0.0001</td>
<td>100 ± 21</td>
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<tr>
<td><strong>AQP2 25 kDa</strong></td>
<td>100 ± 8</td>
<td>63 ± 12</td>
<td>0.029</td>
<td>100 ± 12</td>
</tr>
</tbody>
</table>
Cell model integrating our findings

**Blood**
- Na⁺ → Pds
- K⁺ → Urine

**Principal cell**
- ATP
- K⁺ → Na⁺
- H₂O
- Na⁺ → Cl⁻
- Cl⁻ → HCO₃⁻
- HCO₃⁻ → Urine

**B- Intercalated cell**
- ATP
- H⁺ → NH₃
- NH₃ → Urine

**A- Intercalated cell**
- ATP
- NH₄⁺ → Urine

**Cortex Medulla**
- Aldosterone
- PGE₂

**Medulla**
- ATP
- K⁺ → Na⁺
- Na⁺ → NH₃
- NH₃ → Urine

**Urine**
Conclusions

• Salt and K losing nephropathy associated with type 1 dRTA are primary consecutive to B-IC dysfunction

• B-IC are not only base secreting cells but are also NaCl absorbing cells

• B-IC are also probably biosensor cells controlling most of CCD functions
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