Renal Acid-Base 1 and 2
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OBJECTIVES:

After studying this lecture, the student should understand:

1. The mechanism for reabsorption of filtered bicarbonate.
2. The factors that change reabsorption of filtered bicarbonate.
3. The mechanism for excreting fixed acid as titratable acid, including the role of amount and pK of urinary buffer.
4. The mechanism for excreting fixed acid as ammonium, including the role of urine pH, serum potassium, and adaptive responses to acidosis.
5. How the excretion of fixed acid is altered in disease states such as diabetic ketoacidosis and chronic renal failure.

OPTIONAL READING:

Physiology, Berne and Levy; Mosby, 2004. Chapter 38

I. INTRODUCTION

As you have seen in the previous lectures, the respiratory system regulates the PCO₂. The kidneys regulate the HCO₃⁻ concentration of arterial blood. HCO₃⁻ is the major buffer base in extracellular fluid. The role of the kidneys in normal acid-base balance is three-fold:

A. Reabsorption of filtered HCO₃⁻. Reabsorption of filtered HCO₃⁻ is essential in order to preserve the extracellular stores of HCO₃⁻. This process occurs without the net secretion of H⁺.

B. Excretion of H⁺ as titratable acid. This process involves net secretion of H⁺ and the synthesis of new HCO₃⁻ within the renal cells. Reabsorption of this newly synthesized HCO₃⁻ helps replenish HCO₃⁻ stores in extracellular fluid. Recall that extracellular HCO₃⁻ is used to buffer the fixed acids resulting from metabolism of proteins and phospholipids. Excretion of H⁺ as titratable acid is limited by the amount of buffer, usually HPO₄²⁻, present in tubular fluid.

C. Excretion of H⁺ as NH₄⁺. This process, like formation of titratable acid, involves net secretion of H⁺ and synthesis of new HCO₃⁻ within renal cells. Reabsorption of this newly synthesized HCO₃⁻ also helps replenish
HCO_3^- stores. Excretion of H^+ as NH_4^+ requires synthesis of NH_3 within renal cells; the potential supply of NH_3 is enormous and therefore so is the potential for H^+ excretion as NH_4^+.

II. REABSORPTION OF FILTERED HCO_3^-

A. How much filtered HCO_3^- is reabsorbed?

In a 70 kg male with a glomerular filtration rate of 180 liters/day and a plasma [HCO_3^-] of 24 mEq/L, the filtered load of HCO_3^- is 4320 mEq/day (GFR x plasma concentration). About 2 mEq/day of HCO_3^- are excreted in urine. Thus, 4318 mEq/day are reabsorbed into blood, well over 99% of the filtered load.

Recall that daily ingestion and metabolism of protein and phospholipid produces fixed acid in the body, either H_2SO_4 or H_3PO_4. These fixed acids are buffered by extracellular HCO_3^-; the resulting CO_2 is excreted via the lungs. This type of buffering consumes some of the HCO_3^- in extracellular fluid. Thus, it is important that virtually all of the filtered HCO_3^- be conserved, i.e. reabsorbed. (How the HCO_3^- is replenished by excretion of H^+ as titratable acid or NH_4^+ will be covered later in the lecture.)
Recall that in normal man, virtually all (99.9%) of filtered HCO$_3^-$ is reabsorbed back into blood by the renal tubular cells. The diagram above shows that, primarily in early proximal tubule, H$^+$ are secreted from the cell across the luminal membrane into tubular fluid. This H$^+$ combines with filtered HCO$_3^-$ to form (briefly) H$_2$CO$_3$. H$_2$CO$_3$ quickly decomposes to H$_2$O and CO$_2$ in the presence of brush border carbonic anhydrase. The CO$_2$ formed is highly lipid soluble and so readily crosses the luminal membrane, recombines with H$_2$O inside the cell to form H$_2$CO$_3$ catalyzed by intracellular carbonic anhydrase. H$_2$CO$_3$ dissociates into H$^+$ and HCO$_3^-$. The HCO$_3^-$ is reabsorbed back into peritubular blood and the H$^+$ is secreted across the luminal membrane to combine with another filtered HCO$_3^-$, starting the cycle over again. The net result is reabsorption of filtered Na$^+$ and HCO$_3^-$ without the net secretion of H$^+$. 
The key points are:

1. For every HCO$_3^-$ reabsorbed, a Na$^+$ ion is reabsorbed along with it. The net result is the reabsorption of NaHCO$_3$. Part of the Na reabsorption, primarily in the early part of the proximal tubule, is linked to HCO$_3^-$ reabsorption.

2. No net secretion of H$^+$ occurs via this mechanism. For every H$^+$ secreted into the lumen, a CO$_2$ is formed in the lumen which diffuses back into the cell to form more H$_2$CO$_3$ and more H$^+$. Net reabsorption of HCO$_3^-$ does occur by this mechanism. Furthermore, the more H$^+$ secreted into the lumen, the more filtered HCO$_3^-$ which is reabsorbed. When all of the filtered HCO$_3^-$ has been reabsorbed, then net secretion of H$^+$ can occur (see below).

3. There is little change in the pH of proximal tubule fluid accompanying the reabsorption of filtered HCO$_3^-$. This observation is consistent with lack of net H$^+$ secretion and the presence of luminal carbonic anhydrase.

C. Influences on reabsorption of filtered HCO$_3^-$. 

1. Filtered HCO$_3^-$ load. As the plasma concentration of HCO$_3^-$ changes, the filtered load (GFR x plasma HCO$_3^-$) changes proportionately. Up to plasma concentrations of 40 mEq/L, the amount reabsorbed equals the amount filtered. Above 40 mEq/L, apparent saturation of reabsorption occurs, as if the process were T$_m$-limited. In metabolic alkalosis when the plasma HCO$_3^-$ concentration and the filtered load of bicarbonate are high, excess HCO$_3^-$ is excreted in the urine; this constitutes an important renal mechanism. In metabolic alkalosis, H$^+$ concentration is low, both extracellular and intracellular, thus diminishing the H$^+$ available for secretion into luminal fluid; without secreted H$^+$, the filtered HCO$_3^-$ will not be reabsorbed, but rather excreted in urine.

2. Plasma Cl$^-$ concentration. There is usually a reciprocal relationship between plasma Cl$^-$ concentration and plasma HCO$_3^-$ concentration to maintain charge balance. Thus if plasma Cl$^-$ goes up, plasma HCO$_3^-$ goes down, the filtered load of HCO$_3^-$ goes down, and the amount of HCO$_3^-$ reabsorbed goes down.

3. Extracellular fluid volume. The effect of extracellular volume on NaHCO$_3$ reabsorption is similar to its effect on NaCl reabsorption. Increased extracellular fluid volume decreases NaHCO$_3$ reabsorption in proximal tubule because it increases peritubular capillary
hydrostatic pressure ($P_c$) and decreases peritubular capillary colloid osmotic (oncotic) pressure ($\pi_c$). These changes in Starling forces decrease reabsorption of fluid and electrolytes (NaCl and NaHCO$_3$) from the lateral intracellular space into peritubular capillary blood as shown in the figure below. Conversely, extracellular volume contraction increases proximal HCO$_3^-$ reabsorption because of decreased peritubular capillary hydrostatic pressure and increased peritubular $\pi_c$.

4. **Angiotensin II.** Increased levels of angiotensin II, as would be seen in extracellular fluid volume contraction, stimulates Na$^+$-H$^+$ exchange and therefore increases reabsorption of filtered HCO$_3^-$. This mechanism adds to the direct effects of volume contraction, via Starling forces, described above. Together, the effects of Starling forces and increased angiotensin II in volume contraction leads to a condition called contraction alkalosis, i.e., metabolic alkalosis secondary to ECF volume contraction.
5. **Arterial pCO$_2$.** Increased arterial pCO$_2$ (as in respiratory acidosis) increases filtered HCO$_3^-$ reabsorption by increasing the supply of CO$_2$ to the cells. Remember that CO$_2$ supplies the H$^+$ for secretion. Conversely, decreased arterial pCO$_2$ (as in respiratory alkalosis) decreases filtered HCO$_3^-$ reabsorption because the CO$_2$ supply to the cells is diminished and less H$^+$ is available for secretion. These mechanisms constitute the renal compensations for the respiratory acid-base disturbances and help to restore the arterial pH toward normal.

**Respiratory acidosis:**

\[ \text{↑ PCO}_2 \rightarrow \text{↑ HCO}_3^- \text{ reabsorption} \rightarrow \text{↑ plasma HCO}_3^- \rightarrow \text{↑ arterial pH toward normal} \]

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### III. EXCRETION OF H$^+$ AS TITRATABLE ACID.

A. **Mechanism of formation and excretion of titratable acid.** In this process, HPO$_4^{2-}$ present in the glomerular filtrate is converted to H$_2$PO$_4^-$ by the secretion of H$^+$ into the tubular fluid. The figure below illustrates the process.

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![Diagram of titratable acid excretion](image-url)
Formation of titratable acid occurs in all nephron segments. It depends upon intracellular (but not luminal) carbonic anhydrase. CO₂ is provided to the renal cells either from metabolism or from peritubular blood. CO₂ reacts with H₂O within the cell to form H₂CO₃ and then generates H⁺ and HCO₃⁻. The HCO₃⁻ is reabsorbed into peritubular blood. The H⁺ is secreted into the lumen and combines with filtered HPO₄²⁻ to form H₂PO₄⁻ which is then excreted in the urine. In contrast to the mechanism for reabsorption of filtered HCO₃⁻, net secretion of H⁺ occurs; furthermore the HCO₃⁻ reabsorbed has been synthesized within the renal cells to replenish extracellular HCO₃⁻ stores (depleted from buffering fixed acids).

B. Influences on excretion of titratable acid.

1. Availability of urinary buffers (i.e. HPO₄²⁻). The more HPO₄²⁻ in tubular fluid, the greater the excretion of H⁺ as titratable acid. Phosphate is filtered mainly in the HPO₄²⁻ form because the pH of extracellular fluid is 7.4. i.e.:

\[
\text{pH} = \text{pK'} + \log \frac{\text{HPO}_4^{2-}}{\text{H}_3\text{PO}_4}
\]

\[
7.4 = 6.8 + \log \frac{\text{HPO}_4^{2-}}{\text{H}_3\text{PO}_4}
\]

\[
\text{HPO}_4^{2-} = 4 \quad \frac{\text{H}_3\text{PO}_4}{\text{H}_2\text{PO}_4^-}
\]

As H⁺ are secreted, they titrate HPO₄²⁻ to form H₂PO₄⁻. The minimum urinary pH is 4.4 because distal tubule and collecting duct cells cannot secrete H⁺ against a gradient of more than 1:1000 (3 pH units). When the minimum urine pH is reached, phosphate will exist primarily in the H₂PO₄⁻ form, i.e.:

\[
\text{pH} = \text{pK'} + \log \frac{\text{HPO}_4^{2-}}{\text{H}_2\text{PO}_4^-}
\]

\[
4.4 = 6.8 + \log \frac{\text{HPO}_4^{2-}}{\text{H}_3\text{PO}_4}
\]

\[
\text{HPO}_4^{2-} = 0.004 \quad \frac{\text{H}_3\text{PO}_4}{\text{H}_2\text{PO}_4^-}
\]

Once all phosphate is in the H₂PO₄⁻ form and there is no more HPO₄²⁻ to be titrated, any further H⁺ secretion would cause a drastic fall in urine pH below 4.4 and this is prohibited. Thus, the only way to excrete more H⁺ as titratable acid is to provide more HPO₄²⁻ in the glomerular filtrate.
2. **pK' of the urinary buffer.** A urinary buffer is most effective when the pH of the tubular fluid is within 1.0 unit of the buffer's pk'. The figure above shows that the phosphate buffer pair with a pK' of 6.8 is a more effective urinary buffer than creatinine with a pK' of 4.97. These experiments were done by Dr. Robert Pitts, a world-renowned renal physiologist, on himself. The shaded area under each titration curve shows the total amount of H\(^+\) that is secreted into tubular fluid between the glomerular filtrate (pH 7.4) and the final urine (pH 4.4). Phosphate, with its pK of 6.8, is nearly ideal as a urinary buffer: the linear range of the titration curve overlaps almost perfectly with the range of urinary pH from 7.4 (glomerular filtrate) to 4.4 (minimum pH of final urine). Contrast creatinine, with its pK of 5.0. The linear, or buffering, range of the creatinine curve lies between pH 6 and pH 4. Very little H\(^+\) can be added to urine before urine pH falls to the minimum value of 4.4.

IV. **EXCRETION OF H\(^+\) AS NH\(_4\)\(^+\)**

The second route for excretion of H\(^+\) is NH\(_4\)\(^+\) (60% of total H\(^+\) excretion). This second route is vitally important, since excretion of H\(^+\) as titratable acid is limited by the amount of urinary buffer; as soon as all urinary buffers (usually phosphate) are titrated by H\(^+\) and converted to their HA forms, then urine pH would fall to 4.4 and H\(^+\) secretion would cease. The additional mechanism is the titration of NH\(_3\) to NH\(_4\)\(^+\) by secreted H\(^+\).
Excretion of H\(^+\) as NH\(_4\)\(^+\) involves the proximal tubule, the thick ascending limb of Henle, and the intercalated cells of collecting duct and occurs in the following steps:

1. In the proximal tubule, NH\(_3\) is synthesized from glutamine and is converted to NH\(_4\)\(^+\) inside the cells. NH\(_4\)\(^+\) is secreted into the lumen by substituting for H\(^+\) on the Na\(^+\)-H\(^+\) exchanger. For every NH\(_4\)\(^+\) secreted, one "new" HCO\(_3\)\(^-\) is reabsorbed. The NH\(_4\)\(^+\) travels down the nephron.
2. Some of the NH$_4^+$ secreted by the proximal tubule is excreted in urine. The rest of the NH$_4^+$ substitutes for K$^+$ on the Na$^+$-K$^+$-2Cl$^-$ cotransporter of thick ascending limb and is added to the solutes of the medullary interstitium (not shown in figure above; see countercurrent multiplication). In interstitial fluid, it is present in both NH$_3$ and NH$_4^+$ forms, but primarily as NH$_4^+$ (pK is 9.2, so at pH 7.4, most would be in HA or NH$_4^+$ form).

3. The intercalated cells of collecting duct have H$^+$ ATPase and H$^+$-K$^+$ ATPase in the luminal membrane; both transporters secrete H$^+$ into the lumen of the collecting duct. Then, NH$_3$ (which is highly lipid-soluble) diffuses from the medullary interstitium into the lumen, combines with the secreted H$^+$ to form NH$_4^+$, and is excreted. This process is called diffusion trapping because the lipid-soluble form (NH$_3$) diffuses, is converted to a water-soluble form (NH$_4^+$), which is trapped and then excreted. Note that the lower the NH$_3$ concentration in the lumen, the more favorable the gradient for NH$_3$ diffusion from the medullary interstitium and the more H$^+$ excreted as NH$_4^+$. This point is illustrated in the following calculation based on a urine pH of 7.0:

\[
\text{pH} = \text{pK} + \log \frac{\text{NH}_3}{\text{NH}_4^+} \\
7.0 = 9.2 + \log \frac{\text{NH}_3}{\text{NH}_4^+} \\
\frac{\text{NH}_3}{\text{NH}_4^+} = .006 \\
\text{or} \\
\frac{\text{NH}_4^+}{\text{NH}_3} = 167
\]

In words, at a urine pH of 7.0, there are 167 NH$_4^+$ for every NH$_3$; thus, the luminal concentration of NH$_3$ remains low, maintaining a gradient for NH$_3$ diffusion.

B. **Effect of urine pH on NH$_4^+$ excretion**

The lower the urine pH (of course, with a minimum of 4.4), the more H$^+$ excreted as NH$_4^+$. Why is this? The answer lies in the effect of urine pH on the relative concentrations of NH$_3$ and NH$_4^+$ that we illustrated in the calculation above. If urine pH is 5 instead of 7 (i.e., more acidic), even more is in the NH$_4^+$ form and even less as NH$_3$, making an even more favorable gradient for NH$_3$ diffusion.
C. Adaptive increase in NH$_3$ synthesis in acidosis

Metabolic and respiratory acidosis both produce an adaptive increase in ammoniagenesis after a few days (chronic acidosis). Presumably, there is an associated decrease in intracellular pH that induces key enzymes in the glutamine to NH$_3$ pathway. As a result, more H$^+$ can be excreted as NH$_4^+$ in chronic acidosis than in acute acidosis.

D. Effect of blood [K$^+$] on NH$_3$ synthesis

Plasma K$^+$ concentration also alters the rate of ammoniagenesis and, therefore, the excretion of H$^+$ as NH$_4^+$ concentration. Hyperkalemia inhibits NH$_3$ synthesis and decreases H$^+$ excretion. Hypokalemia increases NH$_3$ synthesis and increases H$^+$ secretion. The effects of plasma K$^+$ may be mediated by H$^+$ and K$^+$ exchange across cell membranes and, therefore, actually may be mediated by intracellular pH. (For example, in hyperkalemia, K$^+$ enters the cells and H$^+$ leaves, increasing intracellular pH and inhibiting NH$_3$ synthesis.)

V. RELATIVE RATES OF EXCRETION OF H$^+$ AS TITRATABLE ACID AND NH$_4^+$ AND ADAPTATION TO METABOLIC ACIDOSIS

<table>
<thead>
<tr>
<th>Condition</th>
<th>mEq of H$^+$ excreted per day</th>
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<tbody>
<tr>
<td></td>
<td>NH$_4^+$</td>
</tr>
<tr>
<td>1. Normal</td>
<td>30 - 50</td>
</tr>
<tr>
<td>2. Diabetic ketoacidosis</td>
<td>300 - 500</td>
</tr>
<tr>
<td>3. Chronic renal disease</td>
<td>0.5 - 15</td>
</tr>
</tbody>
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1. Normally, more \( H^+ \) are excreted as \( \text{NH}_4^+ \) than as titratable acid, over 2/3 of the total amount.

2. In uncontrolled diabetes mellitus, there is overproduction of the fixed acid, \( \beta\text{-OH butyric acid} \), causing metabolic acidosis. This accounts for the large adaptive increase in \( H^+ \) excretion as \( \text{NH}_4^+ \) (10X the normal rate). There is also elevated excretion of \( H^+ \) as titratable acid, resulting from the increased plasma level and filtered load of \( \beta\text{-OH butyrate} \). \( \beta\text{-OH butyrate} \) actually becomes the primary urinary buffer in diabetic ketoacidosis because of its high urinary concentration, even though its pK' (4.8) is not ideal. Thus the high titratable acid excretion is primarily as \( \beta\text{-OH butyric acid} \). Another example of the trade-off of buffer amount vs pK.

3. In chronic renal disease, the decrease in functioning renal cells causes decreased \( H^+ \) excretion as titratable acid and \( \text{NH}_4^+ \). As a result, the excretion of fixed \( H^+ \) is severely compromised and causes a type of metabolic acidosis (broadly termed renal tubular acidosis).