Ureagenesis: evidence for a lack of hepatic regulation of acid-base equilibrium in humans

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Hosch, Markus, Juergen Muser, Henry N. Hultcr, and Reto Krapf. Ureagenesis: evidence for a lack of hepatic regulation of acid-base equilibrium in humans. Am J Physiol Renal Physiol 286: P94–F99, 2004; 10.1152/ajprenal.00283.2003.—Ureagenesis in the liver consumes up to 1,000 mmol of HCO3-/day in humans as a result of 2NH4+ + 2HCO3- → urea + CO2 + 3H2O. Whether the liver contributes to the regulation of acid-base equilibrium by controlling the rate of ureagenesis and, therefore, HCO3- consumption in response to changes in plasma acidity has not been adequately evaluated in humans. Rates of ureagenesis were measured in eight healthy volunteers during control, chronic metabolic acidosis (induced by oral administration of CaCl2 3.2 mmol/kg body wt-1 day-1 for 11 days), and recovery as well as during bicarbonate infusion (200 mmol over 240 min; acute metabolic alkalosis). Rates of ureagenesis were correlated negatively with plasma HCO3- concentration both during adaption to metabolic acidosis and during the chronic, steady-state phase. Thus ureagenesis, an acidifying process, increased rather than decreased in metabolic acidosis. During bicarbonate infusion, rates of ureagenesis decreased significantly. Thus ureagenesis did not appear to be involved in the regulated elimination of excess HCO3-. The finding of a negative correlation between ureagenesis and plasma HCO3- concentration over a wide range of HCO3- concentrations, altered both chronically and acutely, suggests that the ureagenic process per se is maladaptive for acid-base regulation and that ureagenesis has no discernible homeostatic effect on acid-base equilibrium.

Acidosis;
et acid excretion;
alkalosis

Ureagenesis in the liver consumes up to 1,000 mmol of bicarbonate/day in humans as a result of 2NH4+ + 2HCO3- → urea + CO2 + 3H2O.

The liver is believed to contribute to regulation of acid-base equilibrium by controlling the rate of ureagenesis and, therefore, HCO3- consumption in response to changes in plasma acidity. Accordingly, present teaching attributes increased rates of ureagenesis to the defense against HCO3- overload (metabolic alkalosis) and decreased rates to conservation of HCO3- in the defense against metabolic acidosis (9). This clinicopathophysiological concept challenges the traditional view of regulation of acid excretion by the kidney in the principal, if not exclusive, mechanism regulating acid-base balance (18, 24).

Two pathways compete for hepatic NH4+ elimination: ureagenesis (an HCO3- -consuming process, metabolically equivalent to proton generation) and net glutamine synthesis, which does not consume HCO3- directly.1 Based on extensive in vitro

NH4+ effectively yielding 1 net mmol of alkali/mmol NH4+ produced. Accordingly, the shift of NH4+ as a substrate for hepatic urea synthesis to its alternative usage in glutamine synthesis (sparing glutamate oxidation/HCO3- production) provides no change in HCO3- consumption because both alternatives (ureagenesis and glutamine synthesis) are argued to result in 1 net mmol HCO3- consumed/mmol NH4+ consumed (16). However, the unknown quantity of glutamate flux into oxidation vs. other possible fates for glutamate, under both normal conditions and in acid-base disorders, leaves some doubt as to the net acid-base consequences of hepatic ureagenesis vs. glutamine synthesis as alternative pathways for NH4+ consumption. An additional source of uncertainty for the role of glutamine synthesis is that the glutamine synthesized from lactate as a carbon precursor is proton neutral but is proton generating when derived from glucose (1).

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1When glutamine is produced in the liver from NH4+ and glutamate, the reaction neither produces nor consumes bicarbonate. However, if glutamate, the aminocarboxylate that serves as a glutamine precursor, had proceeded to its alternative fate of complete oxidation, it would have produced 2HCO3- plus and in vivo measurements of ureagenesis in acidic and normal rats, it has been argued that the regulation of acid-base composition is accomplished via control of the rate of ureagenesis (3, 9). In acute metabolic acidosis in rats induced by HCl administration, results have been modestly supportive of the hepatic regulator theory, as both decreases (3, 5) and no change (8, 13) in ureagenesis were reported.

In chronic metabolic acidosis (CMA) in rats, however, the results have been reasonably uniform in support of the hepatic acid-base control theory inasmuch as significant reductions in urea production have been observed consistently (6, 19, 23, 27), although one of the reports found only a transient reduction (19). Hepatic control of acid-base homeostasis may also operate in chronic metabolic alkalosis, as chronic alkal exposure in the walking catfish has been recently reported to greatly increase ureagenesis (25). The in vivo data favoring hepatic control of plasma acid-base composition as mediated by acid-base effects on ureagenesis have been confirmed consistently in both in vitro and perfused liver studies in rats (reviewed in Ref. 19). In four normal human subjects administered 200 mmol/day of HCl for 4 days, a significant reduction in urinary urea excretion was observed on day 4, but interpretation is hampered by a lack of standard metabolic control (10).

A regulated shift from ureagenesis to increased glutaminic glutamine release during an acid load would thus ensure continued elimination of toxic NH4+ while simultaneously decreasing HCO3- consumption and, thereby, conserving HCO3-/base stores. Additional support for this thesis comes from perfused liver studies in rats showing that NH4+ flux through glutamine synthetase is stimulated by a decrease in ambient HCO3- concentration and that further increases in hepatic glutamine release in acidosis may be related to an observed diminished nitrogen flux through the hepatic phosphate-dependent glutaminase pathway (decreased ureagenesis)
Table 1. Effect of CaCl₂ administration on steady-state blood and urinary acid-base composition

<table>
<thead>
<tr>
<th>Study Period</th>
<th>pH&lt;sub&gt;blood&lt;/sub&gt;</th>
<th>HCO&lt;sub&gt;3&lt;/sub&gt;⁻, mmol/l</th>
<th>Paco₂&lt;sub&gt;2&lt;/sub&gt;, mmHg</th>
<th>NAE&lt;sub&gt;ur&lt;/sub&gt;, mmol/24 h</th>
<th>NH₄⁺&lt;sub&gt;ur&lt;/sub&gt;, mmol/24 h</th>
<th>TA&lt;sub&gt;ur&lt;/sub&gt;, mmol/24 h</th>
<th>HCO&lt;sub&gt;3&lt;/sub&gt;⁻&lt;sub&gt;ur&lt;/sub&gt;, mmol/24 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>7.40±0.01</td>
<td>26.5±0.3</td>
<td>44.1±0.6</td>
<td>59.2±3.5</td>
<td>47.2±2.5</td>
<td>12.8±2.2</td>
<td>0.8±0.1</td>
</tr>
<tr>
<td>A1</td>
<td>7.35±0.01*</td>
<td>21.2±0.7*</td>
<td>39.0±0.8*</td>
<td>175.4±9.7*</td>
<td>155.7±10.4*</td>
<td>19.7±1.2*</td>
<td>0.0±0.0*</td>
</tr>
<tr>
<td>A2</td>
<td>7.33±0.01*</td>
<td>19.8±0.4*</td>
<td>37.5±0.8*</td>
<td>186.3±4.5*</td>
<td>174.0±4.6*</td>
<td>12.2±1.2</td>
<td>0.0±0.0*</td>
</tr>
</tbody>
</table>

Values are means ± SE (n=8) of the last 2 days in each period. CaCl₂ dosage was 3.2 mmol/kg body wt (BW) -1 day⁻¹. A1 and A2, acidosis periods 1 (oral phosphate intake 0.65 mmol/kg BW⁻¹ day⁻¹, 6 days) and 2 (oral phosphate intake 0.12 mmol/kg BW⁻¹ day⁻¹, 5 days), respectively; NAE, renal net acid excretion; u, urinary; TA, urinary titratable acid excretion. *P < 0.5.

Table 2. Effect of CaCl₂ on steady-state plasma electrolytes, creatinine clearance, and body weight

<table>
<thead>
<tr>
<th>Study Period</th>
<th>Na⁺, mmol/l</th>
<th>K⁺, mmol/l</th>
<th>Cl⁻, mmol/l</th>
<th>Unmeasured Anions, mmol/l</th>
<th>Ca²⁺, mmol/l</th>
<th>PO₄&lt;sup&gt;3-&lt;/sup&gt;, mmol/l</th>
<th>Mg²⁺, mmol/l</th>
<th>Creatinine Clearance, ml/min</th>
<th>BW, kg</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>137.8±0.6</td>
<td>4.0±0.1</td>
<td>107.8±1.3</td>
<td>7.5±1.6</td>
<td>1.23±0.02</td>
<td>1.20±0.07</td>
<td>0.98±0.02</td>
<td>139.2±5.0</td>
<td>71.1±2.8</td>
</tr>
<tr>
<td>A1</td>
<td>136.3±0.7</td>
<td>3.8±0.1</td>
<td>111.2±1.1*</td>
<td>7.7±1.8</td>
<td>1.27±0.00*</td>
<td>1.05±0.06</td>
<td>0.74±0.02*</td>
<td>147.0±6.7</td>
<td>70.1±2.7</td>
</tr>
<tr>
<td>A2</td>
<td>138.0±0.6</td>
<td>3.8±0.1</td>
<td>111.9±1.0*</td>
<td>10.1±1.3</td>
<td>1.26±0.02*</td>
<td>1.27±0.07</td>
<td>0.87±0.02</td>
<td>151.8±6.9</td>
<td>69.9±2.8</td>
</tr>
</tbody>
</table>

Values are the means ± SE (n=8) of the last 2 days in each period. CaCl₂ dosage was 3.2 mmol/kg BW⁻¹ day⁻¹. Unmeasured anions were calculated as the difference of the plasma concentrations of (Na⁺ + K⁺) – (Cl⁻ + HCO₃⁻). *P < 0.05.

Table 3. Effect of CaCl₂ on steady-state urinary electrolyte excretion

<table>
<thead>
<tr>
<th>Study Period</th>
<th>Na⁺, mmol/24 h</th>
<th>K⁺, mmol/24 h</th>
<th>Cl⁻, mmol/24 h</th>
<th>Unmeasured Anions, mmol/l</th>
<th>Ca²⁺, mmol/24 h</th>
<th>PO₄&lt;sup&gt;3-&lt;/sup&gt;, mmol/24 h</th>
<th>Mg²⁺, mmol/24 h</th>
<th>Creatinine Clearance, ml/min</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>226.6±13.7</td>
<td>50.3±2.8</td>
<td>217.6±12.9</td>
<td>49.8±15.6</td>
<td>4.7±1.1</td>
<td>37.8±5.0</td>
<td>4.9±1.1</td>
<td></td>
</tr>
<tr>
<td>A1</td>
<td>203.8±23.5</td>
<td>61.4±4.3*</td>
<td>372.8±32.1*</td>
<td>19.7±11.0</td>
<td>18.1±3.9*</td>
<td>23.9±5.2*</td>
<td>4.0±0.8</td>
<td></td>
</tr>
<tr>
<td>A2</td>
<td>182.8±17.9*</td>
<td>62.0±3.3*</td>
<td>380.8±20.8*</td>
<td>18.1±9.2*</td>
<td>19.8±3.4*</td>
<td>14.4±4.6*</td>
<td>4.2±0.7</td>
<td></td>
</tr>
</tbody>
</table>

Values are means ± SE. CaCl₂ dosage was 3.2 mmol/kg BW⁻¹ day⁻¹. *P < 0.05.

Table 4. Effect of CaCl₂ on stool electrolyte and nitrogen excretion

<table>
<thead>
<tr>
<th>Study Period</th>
<th>Na⁺, mmol/24 h</th>
<th>K⁺, mmol/24 h</th>
<th>Cl⁻, mmol/24 h</th>
<th>Nitrogen, mmol/24 h</th>
<th>Ca²⁺, mmol/24 h</th>
<th>PO₄&lt;sup&gt;3-&lt;/sup&gt;, mmol/24 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>6.4±1.0</td>
<td>27.3±1.7</td>
<td>3.4±2.7</td>
<td>117.1±5.2</td>
<td>2.9±1.3</td>
<td>3.7±1.0</td>
</tr>
<tr>
<td>A1</td>
<td>0.3±0.9*</td>
<td>20.6±1.0*</td>
<td>3.0±1.4</td>
<td>80.0±4.5*</td>
<td>4.6±1.4*</td>
<td>0.4±0.2*</td>
</tr>
<tr>
<td>A2</td>
<td>1.1±0.8*</td>
<td>20.2±1.5*</td>
<td>3.3±1.6</td>
<td>84.1±5.6*</td>
<td>5.1±1.2*</td>
<td>0.5±0.5*</td>
</tr>
</tbody>
</table>

Values are means ± SE (n=8) of the last 2 days (urine) or means per day (stool) of each period. CaCl₂ dosage was 3.2 mmol/kg BW⁻¹ day⁻¹. Urinary unmeasured anions were calculated as (Na⁺ + K⁺ + Ca²⁺ + Mg²⁺) – (Cl⁻ + HCO₃⁻ + phosphate anion). Phosphate anion equivalency was calculated from the corrected pK and urine phosphoric concentration. *P < 0.05.

Table 5. Effect of CaCl₂ administration on nitrogen-containing plasma metabolites

<table>
<thead>
<tr>
<th>Study Period</th>
<th>Glutamine, μmol/l</th>
<th>Glutamate, μmol/l</th>
<th>NH₄⁺&lt;sub&gt;urea&lt;/sub&gt;, mmol/l</th>
<th>Creatinine, μmol/l</th>
<th>Urea, mmol/l</th>
<th>Total Body Urea, mmol</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>462±27</td>
<td>68±8</td>
<td>48±4</td>
<td>77±3</td>
<td>307±15</td>
<td>3.2±0.1</td>
</tr>
<tr>
<td>A1</td>
<td>440±29</td>
<td>70±8</td>
<td>42±4</td>
<td>72±4</td>
<td>309±16</td>
<td>2.9±0.1</td>
</tr>
<tr>
<td>A2</td>
<td>434±38</td>
<td>69±8</td>
<td>44±5</td>
<td>70±2</td>
<td>300±17</td>
<td>2.8±0.1</td>
</tr>
</tbody>
</table>

Values are means ± SE. CaCl₂ dosage was 3.2 mmol/kg BW⁻¹ day⁻¹.

Table 6. Effect of CaCl₂ administration on nitrogen-containing metabolites in urine and stool

<table>
<thead>
<tr>
<th>Study Period</th>
<th>Glutamine&lt;sub&gt;ur&lt;/sub&gt;, μmol/24 h</th>
<th>Glutamate&lt;sub&gt;ur&lt;/sub&gt;, μmol/24 h</th>
<th>NH₄⁺&lt;sub&gt;urea&lt;/sub&gt;, mmol/24 h</th>
<th>Urea&lt;sub&gt;ur&lt;/sub&gt;, mmol/24 h</th>
<th>Urea Production Rate, mmol/24 h</th>
<th>Creatinine&lt;sub&gt;ur&lt;/sub&gt;, mmol/24 h</th>
<th>Uric Acid&lt;sub&gt;ur&lt;/sub&gt;, mmol/24 h</th>
<th>Nitrogen&lt;sub&gt;ur&lt;/sub&gt;, mmol/24 h</th>
</tr>
</thead>
<tbody>
<tr>
<td>Control</td>
<td>580±64</td>
<td>219±11</td>
<td>43.6±2.5</td>
<td>319.5±13.9</td>
<td>312±13</td>
<td>15.4±0.8</td>
<td>3.5±0.3</td>
<td>117.1±5.2</td>
</tr>
<tr>
<td>A1</td>
<td>331±45*</td>
<td>183±16*</td>
<td>152.5±9.6*</td>
<td>347.8±10.6*</td>
<td>340±10*</td>
<td>15.1±0.7</td>
<td>3.1±0.2</td>
<td>80.0±4.5*</td>
</tr>
<tr>
<td>A2</td>
<td>326±49*</td>
<td>170±13*</td>
<td>169.0±5.5*</td>
<td>344.2±14.1*</td>
<td>349±14*</td>
<td>15.3±0.7</td>
<td>3.2±0.3</td>
<td>84.1±5.6*</td>
</tr>
</tbody>
</table>

Values are the means ± SE (n=8) of the last 2 days (urine) or means per day (stool; s) of each period. *P < 0.05 from control. Urea production rate reflects daily appearance in estimated total BW (largely excreted as urea ura) with adjustment for change in total BW urea pool content. Total body urea is body urea content estimated from plasma urea and total BW.
The findings of both increased flux in the glutamine synthetase pathway and decreased flux in the glutaminase pathway in perfused liver have been confirmed with detailed in vitro flux measurements in incubated liver cells from rats with CMA (22). In vivo data in rats with either acute metabolic acidosis or CMA, however, have not confirmed acidosis-induced increases in hepatic glutamine release (5, 6).

The purpose of the present studies was, therefore, to evaluate the direction and magnitude of changes in the rates of ureagenesis in response to decreases (acutely and chronically) and increases in plasma \( \text{HCO}_3^- \) concentration. If ureagenesis contributes to regulation of acid-base equilibrium in humans, a direct correlation among rates of ureagenesis and plasma \( \text{HCO}_3^- \) concentration (or body base content in the steady state) would be expected.

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**METHODS**

Eight healthy male volunteers (71.1 ± 7.9 kg, aged 24–26 yr) were studied under metabolic balance conditions. The metabolic diet provided [per kilogram body weight (BW) and day] 1.8 mmol sodium, 1.05 mmol potassium, 45 ml water, 0.84 g protein, and 36 kcal. CMA was induced by oral administration of 3.2 mmol of \( \text{CaCl}_2 \) kg \( \text{BW}^{-1} \cdot \text{day}^{-1} \) dissolved in water (flavored with strawberry syrup) and ingested in six divided doses. The metabolic diet was supplemented with neutral oral sodium phosphate (0.53 mmol \( \text{kg}^{-1} \cdot \text{day}^{-1} \) phosphate) during control and the first acidosis period (A1), yielding a total phosphate content of 0.65 mmol \( \text{kg}^{-1} \cdot \text{day}^{-1} \) (20.2 mg \( \text{kg}^{-1} \cdot \text{day}^{-1} \)). During the second acidosis period (A2) and during recovery, the sodium phosphate supplement was removed to provide a low-normal daily phosphate content of 0.12 mmol \( \text{kg}^{-1} \cdot \text{day}^{-1} \). The oral phosphate supplement was administered between \( \text{CaCl}_2 \) doses. Oral administration of \( \text{CaCl}_2 \) induces metabolic acidosis by genera-
tion of poorly absorbable CaCO₃ complexes in the intestine, effectively interrupting reabsorption of luminal HCO₃⁻/H₁₇O₂⁻ of pancreatic origin (15).

Arterialized venous blood TCO₂ and pH analysis, determination of plasma and urine electrolytes, and analysis of renal acid excretion and creatinine were performed as previously described (17). Plasma ammonia, glutamine, glutamate, and urea analyses were performed by HPLC after automated precolumn separation (26). Determination of nitrogen in stool was performed with the Kjeldahl method (14). Metabolic period demarcation in stool production was by radiographic identification of orally ingested pellets present in stool (7).

Urea was assumed to be distributed uniformly in total body water. Water space was assumed to be 65% of BW (kg). The “urea production rate” was calculated as the 24-h urinary excretion rate plus the change in urea pool content. The change in urea pool was computed as plasma urea concentration × 0.65 BW (kg) at the beginning − plasma urea concentration × 0.65 BW (kg) at the end of the 24-h sampling periods. The term urea production rate reflects net production or appearance of urea in extracellular fluid and, when used to compute values on a daily basis, is arithmetically similar to urinary excretion, being adjusted only for daily variation in total body water urea pool size.

Values are given as means ± SE. Statistical analysis was performed by ANOVA for repeated measurements.

RESULTS

Tables 1 and 2 illustrate that in the eight healthy human subjects studied, a chronic exogenous acid load administered by CaCl₂ ingestion (3.2 mmol·kg BW⁻¹·day⁻¹) resulted in chronic, hyperchloremic metabolic acidosis of moderate severity, a mean decrease in plasma HCO₃⁻ concentration ([HCO₃⁻]) by 6.8 ± 0.8 mmol/l (P < 0.001). A large increase in renal net acid excretion (NAE) was observed (increase from 59 ± 10 to 186 ± 12 mmol/day, P = 0.005). Reduction of phosphate intake during the second phase of acidosis (decrease in phosphate intake from 0.65 in A1 to 0.12 mmol·kg BW⁻¹·day⁻¹ in A2, Table 1) did not result in significant changes in the severity of acidosis or magnitude of NAE. However, the components of NAE were quantitatively altered. The anticipated significant decrease in renal titratable acid excretion during A2 was offset by a significant increase in renal NH₄⁺ excretion.

Tables 3 and 4 show the changes in urine and stool electrolyte excretion. Tables 5 and 6 show that plasma concentrations of the nitrogen-containing substances glutamine, glutamate, NH₄⁺, and urea did not differ significantly from control during acidosis. As expected, due to enhanced renal ammoniagenesis, urinary glutamine and glutamate excretion rates decreased, whereas urinary NH₄⁺ excretion increased in response to CaCl₂ administration.
The subjects were in approximately neutral nitrogen balance during the control period (Tables 3–6); mean daily nitrogen excretion for urine was computed as 44 mmol/day as NH4, 639 mmol/day as urea, 46 mmol/day as creatinine, and 14 mmol/day as uric acid, totaling 743 mmol of urinary nitrogen/day. When mean stool nitrogen (117 mmol/day) is added, total nitrogen excretion averaged 860 mmol/day. Because measured dietary protein intake (0.84 g day as uric acid, totaling 743 mmol of urinary nitrogen/day. the urea production rate, resulting in an increase in plasma [HCO3−] (Fig. 1) with a rapid and sustained, significant decrease in urine production. During a separate HCO3− infusion protocol performed on day 4 of recovery (200 mmol of NaHCO3/study subject infused over 240 min), plasma [HCO3−] increased further from 27.5 ± 0.9 to 32.5 ± 1.4 mmol/l. As illustrated in Fig. 2, the increase in plasma [HCO3−] into the frankly alkalotic range was associated with a further and significant decline in the urea production rate.

Figure 3 depicts the daily changes in urea production rate compared with the original control period in the case of the two acidosis periods and compared with the last 2 acidosis days in the cases of the recovery period. Induction of acidosis increased the urea production rate, resulting in a significant cumulative increase in production by 500 mmol urea over the 11 days of acidosis (CaCl2 ingestion). By the end of period A2, daily nitrogen balance had become significantly negative vs. that of the control period (−123 ± 48 mmol/day, P < 0.025).

DISCUSSION

The novel results of this study are 1) CMA in humans leads to stimulated rates of ureagenesis and 2) ureagenesis is inhibited during acute metabolic alkalosis (bicarbonate infusion); thus 3) rates of ureagenesis in humans are inversely related to plasma HCO3− concentration in both chronic and acute metabolic disturbances of acid-base equilibrium.

Acidosis was induced by oral administration of CaCl2, an agent that interrupts reabsorption of intestinal luminal HCO3− of pancreatic origin (15). This model of CMA was chosen to avoid alterations in nitrogen intake attendant to NH4Cl acidosis and its potential as an ureagenic stimulus. The use of disparate phosphate intakes to modulate urinary phosphate excretion enabled the experimental partitioning of NAE among NH4+ and titratable acid (comprised of H2PO4−) and thus afforded an examination of ureagenesis during experimentally controlled differences in urinary NH4+ excretion as it might impact the partitioning of total nitrogen excretion among NH4+ and urea. As indicated by Figs. 1 and 3, changes in dietary phosphate did not result in significant changes in ureagenesis, despite significant changes in NH4+ and titratable acid (and phosphate) excretion.

During the acidosis periods, the maximum amount of HCO3− consumed (or, equivalently, protons produced) via ingestion of CaCl2 in a 70-kg subject is estimated as 4.928 mmol (assuming complete complexing of HCO3− by the 3.2 mmol CaCl2·kg BW−1·day−1 over 11 days). The surplus ∼500 mmol of urea generated during this time (Fig. 3) necessitated the consumption of an additional 1,000 mmol of HCO3− during that period, a process directionally maladaptive for acid-base homeostasis.

While the directional differences observed in the ureagenic response to CMA in the humans of the present study and those reported previously in rats (6, 19, 23, 27) remain unresolved, the degree of acidosis reported in the rat studies was modest and of lesser magnitude than that in the human data. Future studies exploring CMA in multiple species and over a range of magnitudes will be of great interest.

Thus our results indicate that the rate of ureagenesis in humans is not geared to subserve acid-base homeostasis but is directionally controlled by the need to remove or retain nitrogen (e.g., NH4+) as results from the net effect of ongoing protein anabolism or catabolism dictated by the metabolic state of the organism and independent of metabolic acid-base status. CMA has been reported to result in negative nitrogen balance in humans (4) and confirmed in the current study due to net catabolism of endogenous protein, a process attributable in rats to glucocorticoid-dependent proteolysis via the ubiquitin/proteasome pathway (20). Thus the changes in catabolic nitrogen waste load during changes in acid-base equilibrium exert a dominant role in the regulation of ureagenesis and override any possible counterbalancing effects of increases and decreases in systemic HCO3− content, even under ambient acid-base conditions (acidosis) that have been demonstrated in vitro and in vivo in rats to inhibit ureagenesis (6, 19, 23, 27) and to stimulate nitrogen flux through hepatic glutamine synthetase and increase hepatic glutamine release during liver perfusion (1, 11, 12) and in vitro incubation (22).

In summary, urea production rates in humans are correlated inversely, and not directly as reported in rats, with base content (plasma [HCO3−]) both during acute and chronic alterations. There is thus no discernible role for ureagenesis in the regulation of acid-base equilibrium in humans. The demonstrated increased urea production in response to acid loads in humans, in itself, consumes HCO3− and thus would potentially aggravate acidosis by causing further depletion of the body’s diminished HCO3− content. It is thus likely that other quantitatively important metabolic processes (e.g., oxidation of glutamate or other alkali-generating reactions) operate in CMA to offset the accelerated proton production of ureagenesis. The nature and regulation of these pathways with respect to maintenance or defense of acid-base equilibrium require further investigation.

The finding that the rate of ureagenesis decreased when body and plasma HCO3− content increased (recovery from metabolic acidosis and induction of metabolic alkalosis) provides additional evidence against a role for ureagenesis in the regulated elimination of base and, therefore, in the prevention or attenuation of metabolic acidosis (primary elevation of base content).

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