Fructose is a six-carbon monosaccharide that naturally exists in honey, fruits, and some vegetables. However, the development of high-fructose corn syrup, as a lower-cost sweetener, has led to a stark rise in the rate of its consumption. One estimate suggests that the average daily intake of fructose in the United States increased 32% between 1978 and 2004 (43). Fructose-1-phosphate is cleaved by aldolase B to dihydroxyacetone phosphate, which is metabolized by fructokinase, also known as ketohexokinase (KHK), an enzyme that has been associated with rapid ATP depletion (14).

Fructose-1-phosphate is cleaved by aldolase B to dihydroxyacetone phosphate and glyceraldehyde, which are substrates for triglyceride synthesis or gluconeogenesis, depending on the cell type. Rapid ATP depletion has been demonstrated to increase circulating levels of uric acid, a byproduct of ATP metabolism, in some individuals (5, 10, 62). Hyperuricemia has been associated with several of the pathological effects of fructose (10, 14, 62).

At the whole body level, high consumption of high levels of dietary fructose has been linked to the development of the metabolic syndrome, i.e., a constellation of disorders consistent...
ing of hyperinsulinemia, hypertension, visceral adiposity, and dyslipidemia (9). Some of these effects are simply a manifestation of increased calorie consumption, and, in this respect, any sugar will do. However, in this regard, fructose does not stimulate the release of pancreatic insulin, a major satiety hormone, which then can lead to overeating. While sex differences in these responses have not been extensively studied, one study (26) did show that fructose increased blood pressure and led to metabolic perturbations in male rats but not in female rats.

Sex differences exist in a number of renal activities including, but not limited to, the regulation of transport, including salt, water, and organic anion and cation reabsorption (40, 46, 52); metabolic activities, including the propensity toward oxidative stress (50); endocrine systems, such as the renin-angiotensin-aldosterone system (64) and endothelin (37); and drug disposition and clearance (52). Moreover, there are a number of metabolic differences between the sexes with regard to nutrient storage and utilization in different tissue beds. For example, men have higher rates of gluconeogenesis and women have higher rates of fatty acid oxidation. Men generally have a higher respiratory quotient and rely on carbohydrates preferentially as an energy source (51). Therefore, it would not be unexpected if renal responses to dietary fructose vary. Indeed, most studies examining the effects of fructose on the kidney have been done in male animals, in general, because these responses have not been extensively studied, one study (26) found that fructose caused increased caloric consumption, and, in this respect, any sugar will do. However, in this regard, fructose does not stimulate the release of pancreatic insulin, a major satiety hormone, which then can lead to overeating. While sex differences in these responses have not been extensively studied, one study (26) did show that fructose increased blood pressure and led to metabolic perturbations in male rats but not in female rats.

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Materials and Methods

Experimental animals. Mice (C57BL/6/129/SV mixed genetic background) were bred and raised at Georgetown University under approved protocols within the established guidelines of the Institutional Animal Care and Use Committee of Georgetown University. Two-month-old male and female mice (n = 6 mice per sex per treatment) were randomized to receive control (TD01457, Teklad) or high-fructose (65% by dry weight, TD01458, Teklad) diet as pelleted treatment. Mice were randomized to receive control (TD01457, Teklad) or high-fructose (65% by dry weight, TD01458, Teklad) diet as pelleted treatment. The area under the curve for glucose tolerance was calculated for each tubule sampled.

Glucose tolerance testing. Near the end of the 3-mo period, glucose tolerance was measured in mice. Mice were fasted for 5 h and then injected intraperitoneally with filter-sterilized 20% dextrose solution (2 g/kg body wt). Tail vein blood glucose was measured at 0, 15, 30, 45, 60, and 90 min. The area under the curve for glucose tolerance was calculated based on Tai’s model (56).

Histochemical analyses. The right kidney was embedded in paraffin blocks and sectioned by the Histology Core (Lombardi Cancer Center, Georgetown University). Sections from 3 mice/group were stained with Masson’s trichrome and analyzed for histological evidence of pathology. Sections were imaged with a Zeiss Axiosport 410 inverted microscope. Sections from 4 mice/group were semiquantitatively analyzed for GLUT5 subcellular localization using immunoperoxidase-based staining. Heat-induced target retrieval was performed on slides using citrate buffer (pH 6, DakoCytomation, Carpinteria, CA) to unmask antigenic sites. Endogenous peroxidase activity was removed by incubation with 3% H2O2 for 10 min and blockade with avidin/biotin (Vector) and 10% normal goat serum. Sections were incubated with GLUT5 primary antibody (PA1737, Boster Immunoleader, 1:2,000) overnight at 4°C. Secondary antibody was biotinylated goat anti-rabbit antibody (BA-1000, Vector Labs, 1:1,000).

Statistics. All data were analyzed using SigmaPlot (Systat Software, version 10, Evanston, IL). Two-way ANOVA was used to determine differences due to the main factors, sex, diet, and their interactions for all variables. One-way ANOVA followed by a multiple-comparisons test or unpaired r-tests were used to determine differences between individual pairs of means. P values of <0.05 were considered to be significant.
RESULTS

Metabolic effects of fructose feeding. Body weight and weight gain (Table 1) were not affected by fructose feeding; however, male mice of both treatments were ~35% heavier and gained twice as much weight. Male mice had a significantly reduced ability to rapidly clear a bolus of intraperitoneal glucose (glucose tolerance) compared with female mice. Surprisingly, fructose feeding improved this ability in male mice. Male mice also had significantly increased plasma insulin, and there was a significant interactive term in that female mice showed a slight increase in plasma insulin with fructose and male mice showed a decrease. Plasma aldosterone was not significantly affected by sex or treatment.

Hypertrophy of the kidney and osmotic diuresis. Fructose feeding resulted in significantly increased kidney wet weight (Fig. 1A). The increase was ~19% and 10% in male and female mice relative to sex-respective control mice. Absolute weights of the kidneys were heavier in male mice (not shown), but no sex differences were observed when kidney weight was normalized to body weight. Urine volume (Fig. 1B) was also significantly higher in fructose-fed mice of both sexes (increases of 77% and 327% in male and female mice, respectively). Urine osmolality, however, was not significantly lower in fructose-fed mice (Table 2). This resulted in a large and significant increase in total osmoles excreted in fructose-fed mice (Fig. 1C). Moreover, there was no sex difference in this parameter when normalized to body weight. Dietary fructose led to a large significant increase in urinary fructose (Table 2). Fructose was increased >100-fold in male mice and 40-fold in female mice. Interestingly, male control mice had significantly lower (less than half) urine fructose compared with female control mice. Gross differences in collagen deposition in the cortex were evaluated by Masson’s trichrome staining (Fig. 2). In general, no obvious differences were observed as a result of diet or sex.

Uric acid metabolism. Uric acid metabolism was evaluated as a determinant of ATP depletion. Urine uric acid was significantly increased by fructose feeding (>100%) in both male and female mice, with no sex differences (Fig. 3A). Surprisingly, uric acid concentrations in plasma were highest in male control mice and reduced significantly in both sexes by fructose feeding (Fig. 3B). Uric acid clearance was highest in female mice fed fructose, with an increase of >500% compared with female control mice (Fig. 3C).

Urine electrolytes. Dietary fructose led to a significant increase in the excretion of Na⁺, K⁺, and Cl⁻ in urine (Table 2). There were no sex differences in this excretion (when normalized to body weight). The ratio of electrolytes in urine was also evaluated as an index of distal tubular electrolyte homeostasis. Fructose did not significantly affect any of these ratios (as assessed by two-way ANOVA); however, female mice had a lower ratio of K⁺ to Cl⁻ in urine, and this difference was attenuated in fructose-fed mice. The fractional excretion of Na⁺, K⁺, and Cl⁻ is shown in Fig. 4; mice are plotted individually to show increased variability in fructose-fed animals. Fructose increased the fraction excretion of all three electrolytes. This response was enhanced in male mice (highly significant interactive term). Differences in the fraction excretion of electrolytes between sexes were primarily driven by a fall in urine creatinine in male fructose-fed mice.

Plasma electrolytes. Plasma Na⁺, K⁺, and Cl⁻ were all significantly affected by fructose feeding (Fig. 5). Plasma Na⁺ (Fig. 5A) and Cl⁻ (Fig. 5C) were reduced by fructose, but only in female mice (significant interactive term). Plasma K⁺, on the other hand, was increased by fructose in both sexes. The increase was larger in female mice, as female control mice had the lowest plasma K⁺. The ratio of Na⁺ to Cl⁻ was not different between groups, which indicates that these two electrolytes were being regulated in parallel. In contrast, ratios of Na⁺ to K⁺ and K⁺ to Cl⁻ were highly affected by fructose in that fructose reduced the former and increased the latter. There was also a significant interaction for both of these ratios in that the effects were strongest in female mice.

Expression of proteins involved in water balance. Due to evidence of sex differences in plasma electrolytes with fructose feeding (modest hyponatremia in female mice), we tested the cortical expression of two major vasopressin-regulated proteins involved in urine concentrating, i.e., AQP2 in the collecting duct (CD) and NKCC2 in the thick ascending limb (TAL; Fig. 6). Both AQP2 and NKCC2 were more highly expressed in female control mice. Fructose feeding reduced both proteins, and the degree of reduction was greater in female mice.

Expression of proteins involved in fructose metabolism/transport. In contrast, the expression of proteins involved in fructose transport/metabolism or energy homeostasis (Fig. 7)
To determine whether sex or dietary fructose affect the order for luminal-to-interstitial directional transport to occur, the PT requires apical brush-border localization in the first enzyme in fructose metabolism, and the insulin receptor β-subunit were increased by fructose only in male mice (2). However, female control mice had greater cortical expression of GLUT5 compared with female mice. Male mice had substantially greater cortical expression of GLUT5 protein under control and dietary fructose feeding (Fig. 6). Furthermore, whereas the expression of GLUT5 increased twofold in male fructose-fed mice, it was unresponsive in female mice. The increase in renal GLUT5 of other laboratories (1, 2).

DISCUSSION

The consumption of fructose, around the world, has increased substantially in the last three to four decades (8, 9, 53, 57). However, whether or not this represents an inherent health risk and whether certain populations may be more susceptible are still uncertain. Our experiments were aimed to determine whether there were sex differences primarily in the renal-specific responses to fructose feeding in the mouse. We used mixed genetic background (~80% 129/SV and 20% C57Bl6/J). It is important to note that background strain may affect overall results. Our mixed background would be predicted to increase overall variability, and therefore any sex differences found might be considered more robust. Overall, we found a pattern of differences that we interpret as supportive of greater PT metabolism of fructose by male mice and a greater impact, i.e., sensitivity of the distal tubule (TAL and CD) in female mice. We will discuss these findings and interpretation in greater detail below.

Like glucose, fructose has the capacity to be reabsorbed from the filtrate in the PT. Whereas Scl5a2, i.e., Na+-glucose cotransporter 2 (SGLT2), is responsible for the majority of glucose reabsorption in the PT (63), it does not transport fructose (63). In the Scl2 family, Scl2a5 (GLUT5) appears to be the major facilitator of fructose reabsorption across the apical membrane of the PT (2). Our Western blot analysis results support our conclusion that male mice have higher total PT cellular levels of GLUT5 protein under control and dietary fructose feeding (Fig. 6). Furthermore, whereas the expression of GLUT5 increased twofold in male fructose-fed mice, it was unresponsive in female mice. The increase in renal GLUT5 with fructose feeding in male mice confirmed the observation of other laboratories (1, 2).

Nevertheless, our immunohistochemical analysis (although clearly not as quantitative) did not show increased density in male animals or due to fructose in either sex. In fact, female appeared to be more sensitive to fructose feeding in male mice. Male mice had substantially greater cortical expression of GLUT5 compared with female mice. GLUT5 expression was increased by ∼50% in male mice by fructose, but female mice did not show sensitivity to fructose feeding with regard to GLUT5 abundance. Similarly, KHK, the first enzyme in fructose metabolism, and the insulin receptor β-subunit were increased by fructose only in male mice (significant interaction).

Subcellular localization of GLUT5. GLUT5 in the S3 segment of the PT requires apical brush-border localization in order for luminal-to-interstitial directional transport to occur (29). To determine whether sex or dietary fructose affect the relative amount of protein in the apical versus basolateral aspects of the cell, we performed semiquantitative immunohistochemistry (Fig. 8). Surprisingly, fructose had no effect in male mice on the average density in the basolateral or apical aspects sampled; however, female control mice had greater apical density (relative to background) compared with fructose-fed mice (Fig. 8B). Figure 8C shows a comparison of the ratio of apical to basolateral staining. There was a clear sex difference in that female mice had a greater ratio regardless of dietary treatment.

Na+-K+-ATPase expression in regions of the kidney. To determine whether electrolyte differences might result from altered driving forces, we evaluated protein levels (Fig. 9) of α1-subunit of Na+-K+-ATPase pump in homogenates from the cortex, inner stripe of the outer medulla, and inner medulla. Female mice had modest but significantly higher levels of the α1-subunit in homogenates from the cortex (primarily PT associated) and inner medulla (CD associated) compared with male mice (two-way ANOVA). In addition, fructose had a tendency to increase expression in female mice only in all three regions, leading to a significant interactive term for homogenates from the inner stripe of the outer medulla (TAL associated) and inner medulla with a strong trend (P = 0.061) in cortex homogenates.
mice (on both diets) had a higher ratio of apical to basolateral staining, potentially indicative of transport potential. The absence of finding an increase in GLUT5 due to fructose feeding with immunohistochemistry in either sex might be explained by hypertrophy of the PT (10% in female mice and 19% in male mice), which may have diluted the signal with immunohistochemistry but would not confound Western blot analysis. Nonetheless, in female mice fed fructose, both apical and basolateral staining, as quantified, was significantly reduced (relative to same-sex control mice). Thus, it appears that female mice either downregulate or at least have attenuated upregulation of GLUT5 expression (relative to male mice) in response to dietary fructose, in agreement with the Western blot analysis results and directional changes in KHK.

Based on the fairly high increase in fructose in urine in fructose-fed mice, it is likely that fructose uptake into PT cells is not nearly as efficient as that of glucose. In support of this, the \( K_m \) of GLUT5 in rat has been reported to be 12.6 mM, similar to the small intestine, and rather high relative to blood levels (0.1–0.3 mM in rats that consumed fructose) (11). A more recent study (15) examining mouse GLUT5 in transfected oocytes found similar kinetics to those of the rat. In comparison, SGLT2 has a \( K_m \) of 1.6 mM for glucose, but blood levels are near 5.5 mM (35). However, the fact that male mice had more GLUT5 and this level was increased by fructose suggests that they had the capacity to adapt in this manner. This may be due to the fact that GLUT5 is an “Sry-regulated” gene (15). Sry is a transcription factor produced from the Y chromosome.

### Table 2. Urine analysis

<table>
<thead>
<tr>
<th>Group</th>
<th>Urine Osmolality, osm·kg·H_2O</th>
<th>Urine Fructose, μmol/day</th>
<th>Urine Na⁺, μmol/day</th>
<th>Urine K⁺, μmol/day</th>
<th>Urine Cl⁻, μmol/day</th>
<th>Ratio of Na⁺ to Cl⁻</th>
<th>Ratio of Na⁺ to K⁺</th>
<th>Ratio of K⁺ to Cl⁻</th>
</tr>
</thead>
<tbody>
<tr>
<td>MC</td>
<td>2.08 ± 0.32</td>
<td>3.14 ± 0.81</td>
<td>54 ± 11</td>
<td>105 ± 18</td>
<td>71 ± 15</td>
<td>0.95 ± 0.25</td>
<td>0.53 ± 0.06</td>
<td>1.72 ± 0.25</td>
</tr>
<tr>
<td>MFr</td>
<td>2.54 ± 0.36</td>
<td>430 ± 132</td>
<td>105 ± 19</td>
<td>240 ± 45</td>
<td>145 ± 27</td>
<td>0.73 ± 0.03</td>
<td>0.44 ± 0.03</td>
<td>1.68 ± 0.10</td>
</tr>
<tr>
<td>FC</td>
<td>2.85 ± 0.18</td>
<td>8.19 ± 1.56*</td>
<td>47 ± 7</td>
<td>68 ± 15</td>
<td>62 ± 9</td>
<td>0.77 ± 0.12</td>
<td>0.62 ± 0.05</td>
<td>1.26 ± 0.05</td>
</tr>
<tr>
<td>FFr</td>
<td>2.84 ± 0.40</td>
<td>334 ± 74</td>
<td>91 ± 12AB</td>
<td>174 ± 23AB</td>
<td>115 ± 14AB</td>
<td>0.79 ± 0.05</td>
<td>0.53 ± 0.05</td>
<td>1.52 ± 0.10AB</td>
</tr>
</tbody>
</table>

**Results of two-way ANOVA (diet × sex) (P values)**

<table>
<thead>
<tr>
<th>Diet</th>
<th>0.51</th>
<th>&lt;0.001†</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sex</td>
<td>0.12</td>
<td>0.29</td>
</tr>
<tr>
<td>Interaction</td>
<td>0.48</td>
<td>0.27</td>
</tr>
</tbody>
</table>

Values are means ± SE; \( n = 6 \) mice/group. Daily excretion of urine fructose, Na⁺, K⁺, and Cl⁻ were normalized to 25 g of body weight. \( ^A,B \)Letters indicate the results of a multiple-comparisons testing after a significant (\( P < 0.05 \)) one-way ANOVA. “A” is equal to “AB” but not “B”. *Significantly different from the MC group by an unpaired t-test. †Significant values (\( P < 0.05 \)) for two-way ANOVA results.
mosome that increases the expression of this transporter, similar to other proteins involved in differentiation of the male sex (55). In a recent study by Aoyama et al. (1), increased outer cortical GLUT5 expression in response to dietary fructose corresponded temporally to increased tubulointerstitial fibrosis in the DBA/2N mouse strain, suggesting that cellular uptake of the sugar was necessary to initiate some modes of renal pathology.

This difference in GLUT5 expression may have also precipitated the ~100% increase in the expression of KHK in male fructose-fed mice. KHK and GLUT5 expression are thought to be limited exclusively to the PT in kidney (18, 19). Thus, while other renal tubule cells may reabsorb filtered fructose or transport it from the basolateral side, it is unclear whether they can metabolize it. PT cells have similarities to hepatic cells, the major metabolic site for absorbed fructose. In that regard, our results do contrast to what has been reported in rats fed fructose, where female rats were found to have a greater upregulation of fructokinase (KHK) in the liver compared with male rats (61). In addition, female rats in that study (6) suffered additional consequences related to diet (impaired glucose tolerance test and elevated plasma insulin) to a greater extent than male rats (61). This did agree with our findings in mice on the whole body impact of dietary fructose, which, we found, were somewhat more apparent in female mice, e.g., increased plasma insulin and a trend for reduced glucose tolerance test.

We also found that male mice had an approximately fourfold increase in the expression of renal cortical insulin receptor (β-subunit) with fructose feeding, whereas female mice started out with higher levels of expression, which did not change with fructose. Whether this increase in receptor number led to greater signaling capacity in these cells or was in response to insulin receptor “resistance” is not known. We have found a

![Figure 3](http://ajprenal.physiology.org/)

**Fig. 3.** Male mice had greater uric acid in plasma. *A*: urine uric acid excretion. *B*: plasma uric acid. *C*: uric acid clearance. Data were normalized to 25 g of body weight. “A” is significantly greater than “B” but not “AB” [results of a multiple-comparisons test after a significant (*P* < 0.05) one-way ANOVA]; two-way ANOVA (sex × diet) results are provided in the graphs.

![Figure 4](http://ajprenal.physiology.org/)

**Fig. 4.** Fractional excretion of Na⁺ (*A*), Cl⁻ (*B*), and K⁺ (*C*) in mice fed control or fructose diets. Each mouse is plotted separately. Fractional excretion of all three electrolytes was increased by fructose feeding, but predominantly in male mice.
number of factors that influence expression of the renal insulin receptor, including obesity and high-fat diets (decrease expression) as well as insulin infusion (increase its expression) (59). Impaired insulin signaling in the PT may be associated with enhanced gluconeogenesis at this site (28).

Nonreabsorbed fructose travels down the tubule lumen, where it can act as an osmolyte, drawing in fluid, leading to osmotic diuresis. We did not find any sex differences in the absolute or body weight-normalized amount of fructose in urine, which was substantial in both sexes (430 μmol or 77 mg/day for male mice) and represented 0.3 kcal (4 kcal/g 0.077 g fructose). Nonetheless, maintenance energy requirements for a lean mouse have been estimated at 124 kcal·kg⁻³/₄·day⁻¹ (39) or 10 kcal/day for a 30-g mouse. Thus, urinary fructose would represent only ~3% of daily energy requirements and should not have substantially affected nutritional status.

Although we did not measure food intake as it is difficult with pelleted diet in chronic studies, we did monitor urine electrolytes as an index of food intake in the chronic state. The diets were formulated to contain the same percentage of these electrolytes. While there were no significant differences in body weight gain between fructose- and control diet-fed mice, urine electrolytes were increased by ~94%, 129%, and 104% for Na⁺, K⁺, and Cl⁻, respectively, in male mice and by 94%, 156%, and 85% in female mice for fructose-fed animals. It is not entirely clear what is driving this increase in urinary electrolytes. The possibilities include 1) increased food consumption due to lower energy bioavailability of fructose and/or increased basal metabolic rate or 2) losses from other tissues beds, such as bone, muscle, and extracellular fluid. Calculation of the fractional excretion of electrolytes revealed a greater response in male mice; however, this sex effect was primarily driven by a fall in urine creatinine in male fructose-fed mice rather than increased urine electrolytes. Why male and not female mice experienced this reduction in urine creatinine with fructose feeding is unclear. Plasma creatinine levels were not significantly different.

The osmotic diuresis produced by fructose may have been the cause of the slight, but significant, disruption in plasma...
electrolyte homeostasis. Overall, fructose increased the ratio of $K^+$ to both $Cl^-$ and $Na^+$ in the plasma, and this effect was significantly greater in female mice. Female mice fed fructose had significantly lower plasma $Na^+$ and $Cl^-$ than all other groups. Both sexes experienced some rise in serum $K^+$. Although the fall in serum $Na^+$ and $Cl^-$ in female mice was modest, it might be exacerbated with aging or comorbidities. Verbalis and colleagues (40) have demonstrated sex differences in hyponatremia, with females being more sensitive. The rise in plasma $K^+$ could be the result of impaired excretion or transcellular shifts in $K^+$ (44). Our urine analysis did not indicate impaired $K^+$ excretion. In fact, $K^+$ showed the greatest percentage increase in excretion with fructose. Similarly, the rise in expression for the $\alpha_1$-subunit of $Na^+-K^+-2Cl^-$ cotransporter (NKCC2) would be predicted to increase $K^+$ excretion in exchange for $Na^+$ resorption at least in the CD. Thus, we can

![Figure 6](http://ajprenal.physiology.org/) Aquaporin (AQP2) and $Na^+-K^+-2Cl^-$ cotransporter (NKCC2) abundance showed greater sensitivity to fructose in female mice. A: Western blots of cortex homogenates (each lane shows a different mouse sample) loaded with equal amounts of protein and probed with antibodies against AQP2, NKCC2, and GAPDH. GAPDH (reprobe) was used to normalize loading. "A" is significantly greater than "B" but not "AB" [results of a multiple-comparisons test after a significant ($P < 0.05$) one-way ANOVA]; two-way ANOVA (sex × diet) results are provided in the graph (B).

![Figure 7](http://ajprenal.physiology.org/) Proteins involved in energy metabolism are upregulated by fructose in male mice. A: Western blots of cortex homogenates (each lane shows a different mouse sample) loaded with equal amounts of protein and probed with antibodies against glucose transporter (GLUT)5, ketohexokinase (KHK), insulin receptor β-subunit (IR-β), and GAPDH. GAPDH (reprobe) was used to normalize loading. "A" is significantly greater than "B" but not "AB" [results of a multiple-comparisons test after a significant ($P < 0.05$) one-way ANOVA]; two-way ANOVA (sex × diet) results are provided in the graph (B).

![Figure 8](http://ajprenal.physiology.org/) GLUT5 immunohistochemistry. A: representative stained sections from the deep cortical region of the kidney from a MC mouse (top left), MFr mouse (top right), FC mouse (bottom left), and FFr mouse (bottom right). Magnification: ×400. B: summary of density semiquantification of “apical” and “basolateral” aspects of tubules sampled (sampling 10 tubules/mouse, n = 4 mice/group). C: summary of average mean ratios of apical to basolateral staining calculated for each tubule. *Significant ($P < 0.05$) difference between control- and fructose-fed mice within the female sex (by unpaired t-test). "A" is significantly greater than "B" [results of a multiple-comparisons test after a significant ($P < 0.05$) one-way ANOVA]; two-way ANOVA (sex × diet) results are provided in the graphs.
assumed that the increase in this subunit (which has been shown to be aldosterone sensitive) (47) may represent an attempt to compensate for hyperkalemia rather than the cause of it. The relative hyponatremia and hypochloremia in female fructose-fed mice were accompanied by a fall in the expression of two major proteins involved in water balance, i.e., AQP2 and NKCC2. We also found a fall in the abundance of these two proteins in our study done in male rats in response to fructose (54). The cause-and-effect relationship between the fall in these proteins and the 237% increase in urine volume in female mice is uncertain. A reduction in the level of these proteins would clearly facilitate water excretion both by affecting urinary concentrating and diluting capacity of the TAL and affecting permeability of the CD. The fall in these proteins would have been predicted to protect mice from more severe hyponatremia (23, 40). It is unclear why these proteins were not downregulated in male mice as well with fructose; however, control levels were lower in male mice. Furthermore, this supports a more distal phenotype in female mice. Another study (33) has suggested that fructose feeding may lead to dehydration and stimulate vasopressin release. Vasopressin has been shown to upregulate the abundance of both AQP2 (20) and NKCC2 (24). Therefore, we do not predict that vasopressin levels were elevated in our study.

High dietary fructose has been associated with the development of the constellation disorder known as metabolic syndrome in humans and animals models, i.e., dyslipidemia, hypertension, visceral adiposity, and hyperinsulinemia (9, 25, 57). One potential contributing mediator to various aspects of metabolic syndrome is hyperuricemia (4, 32, 41, 45). Uric acid is the end product of purine metabolism generated by the action of the enzyme xanthine oxidase (4). We found an interesting phenomenon with regard to uric acid homeostasis in our mice. While urine excretion of uric acid was significantly increased by high-fructose diet, as we predicted, plasma levels were reduced. In fact, male control mice had the highest levels of plasma uric acid. Reduced levels of plasma uric acid have also been found in patients with type 1 diabetes (5, 6). One potential explanation provided is that poorly controlled diabetes resulting in high circulating glucose levels can impair PT function (Fanconi-like syndrome), resulting in reduced serum circulating levels of uric acid, as urate can be reabsorbed at this site. Another explanation could be that high luminal fructose is competitively inhibiting urate reabsorption through transporters such as GLUT9 (Slc2a9) (38). GLUT9 has recently been shown to have the capability to transport urate in addition to glucose and is expressed in the renal PT, along with GLUT5.

In the present study, we did not elect to measure blood pressure. In a previous study in rats (54), we did not find an effect of dietary fructose on blood pressure, similar to other published reports (3, 17, 27). In contrast, some laboratories have observed fairly substantial increases (12, 13, 30, 31) and even a sex difference in this response (26). Because it is likely that this response is very dose/strain/species specific, we did not elect to focus on this aspect in the present report. Moreover, we did not find clear evidence of tubulointerstitial fibrosis of glomerulopathy in our fructose-fed mice. Our mice were not prone to excessive weight gain as a result of the fructose diet, which may have ameliorated many of the metabolic effects. We feel that this is a strength of our study as we did not have to interpret these confounding influences. The lack of severe renal pathology is also consistent with other studies in mice showing only modest changes, which may be strain specific (1).

In summary, fructose feeding resulted in differential responses at the level of the kidney in male versus female mice. Our findings are consistent with more proximal effects in male mice that may arise from increased cellular uptake and metabolism and a more distal phenotype in female mice, with effects on serum electrolytes and expression of transporters/channels that regulate these parameters. Nonetheless, it is not clear whether the changes in female mice represent adaptive, maladaptive, or neutral responses to fructose. Additional studies are warranted to address differences in vulnerabilities to dietary fructose between the sexes and the mechanisms underlying them.

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