Hill's
Transforming Lives

# Activating the gut microbiome helps protect kidney health in cats & dogs with chronic kidney disease

## Clinical Evidence Report

### Key Findings

- Chronic kidney disease (CKD) is associated with an altered intestinal microbiome (dysbiosis), resulting in production of gut-derived uremic toxins that are harmful to the kidneys and have been associated with CKD progression.
- Even without obvious clinical signs in early stages of CKD in cats and dogs, harmful uremic toxins are present.
- Controlling amounts of high-quality/highly digestible dietary protein decreases uremic toxins compared with feeding higher protein in early CKD.
- Feeding a therapeutic renal food (Prescription Diet k/d with ActivBiome+ Kidney Defense) that contains prebiotics and betaine significantly decreases gut-derived uremic toxins and provides other benefits.

A growing body of evidence indicates that chronic kidney disease (CKD) is associated with an altered intestinal microbiome (dysbiosis), which leads to microbial production of uremic toxins that can injure the kidneys and contribute to progression of CKD.[1-6] Clinical signs may be subtle (e.g., weight loss) for years or not observed in cats and dogs with CKD, especially in early stages;[7,8] however, uremic toxins are already being produced.[1-3,9,10] These gut-derived uremic toxins have many negative effects, which may help explain the pathogenesis and clinical consequences of CKD.[11-15]

| Uremic Toxins | Harmful Effects |
|---|---|
| • Advanced glycation end-products<br>• Hippurates (e.g., hippuric acid)<br>• Indoles (e.g., indoxyl sulfate)<br>• Phenols (e.g., p-cresol sulfate)<br>• Polyamines (e.g. putrescine)<br>• Others - homocysteine, TMAO | • Promote oxidative injury, inflammation and fibrosis in renal tubules<br>• Contribute to CKD progression<br>• Impair erythropoietin synthesis<br>• Decrease colonic motility & constipation<br>• Contribute to loss of muscle mass |

In healthy individuals the intestinal barrier (including tight junctions, enterocyte membranes, and mucus layer) prevents translocation of substances and microbes from the intestinal lumen into the bloodstream.[4-6] The intestinal microbiome plays a key role in maintaining barrier function by modulating the immune system and controlling luminal pH, which helps protect against harmful bacterial colonization and dysbiosis. Adequate dietary fibre intake supports growth of beneficial saccharolytic bacteria, which produce helpful compounds (post-biotics such as short-chain fatty acids) that nourish intestinal cells and contribute to a healthy intestinal barrier.

The pathogenesis of dysbiosis in CKD is linked to the bi-directional communication between the kidneys and the intestinal tract (gut-kidney axis) **(Figure 1)**. Potential factors contributing to this dysbiosis include reduced intake of dietary fibres, prolonged colonic transit time (constipation), increased amounts of protein available for digestion by proteolytic bacteria in the colon and changes in the colonic microbiota.[16] Decreased dietary fibre intake favors growth of proteolytic bacteria, which ferment amino acids to produce potentially harmful metabolites including ammonia (which is converted to ammonium hydroxide) and gut-derived uremic toxins. These toxins incite a local inflammatory response and increased intestinal permeability, which allows for translocation of intestinal bacteria and gut-derived uremic toxins into the bloodstream where they can lead to systemic inflammation and renal injury.[16,17]

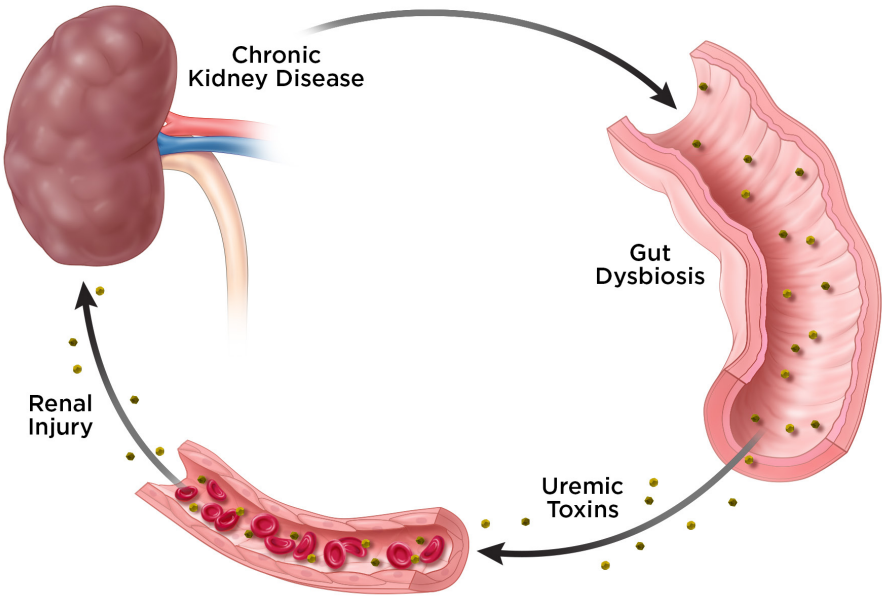


**Figure 1.** There is a bi-drectional relationship between the kidneys and the gut microbiome (gut-kidney axis). CKD is associated with disruption of the gut microbiome (dysbiosis) leading to production of gut-derived uremic toxins, which contribute to progression of CKD and retention of additional uremic toxins that further perpetuates dysbiosis (a vicious cycle).

## New Nutritional Interventions for CKD

Nutritional management is the standard of care for CKD and is the only treatment shown to improve length and quality of life in cats and dogs with CKD.[18-20] The best quality evidence (Grade 1) from randomized controlled clinical trials shows that feeding one therapeutic renal food (Prescription Diet k/d) results in significantly longer survival times and better quality of life for cats and dogs with CKD compared with feeding a typical pet food.[19,20] Regarding lowering uremic toxins, the focus has been on feeding decreased amounts of dietary protein in later stages of CKD. However, due to recent discoveries about harmful effects of uremic toxins in early CKD and the impact of dysbiosis, additional management options are needed. In addition, gut-derived uremic toxins are protein-bound and cannot be effectively eliminated by dialysis or fluid therapy; therefore, novel nutritional options have been evaluated for their effects on decreasing uremic toxin production.[21,22]

At the Hill's Pet Nutrition Center we have conducted research evaluating effects of different nutritional interventions on the intestinal microbiome and subsequent uremic toxin production in healthy cats & dogs and those with CKD.[23-26] Based on these foundational studies, we then evaluated two nutritional approaches: 1) feeding decreased amounts of high-quality/highly digestible protein, so there is less substrate for gut bacteria to produce uremic toxins, and 2) including betaine and prebiotic fibres (short-chain fructooligosaccharide and oat beta glucan) in the food to shift the balance of gut bacteria to those utilizing carbohydrates (saccharolytic) versus those fermenting protein (proteolytic), leading to decreased production of harmful uremic toxins **(Figure 2)**.

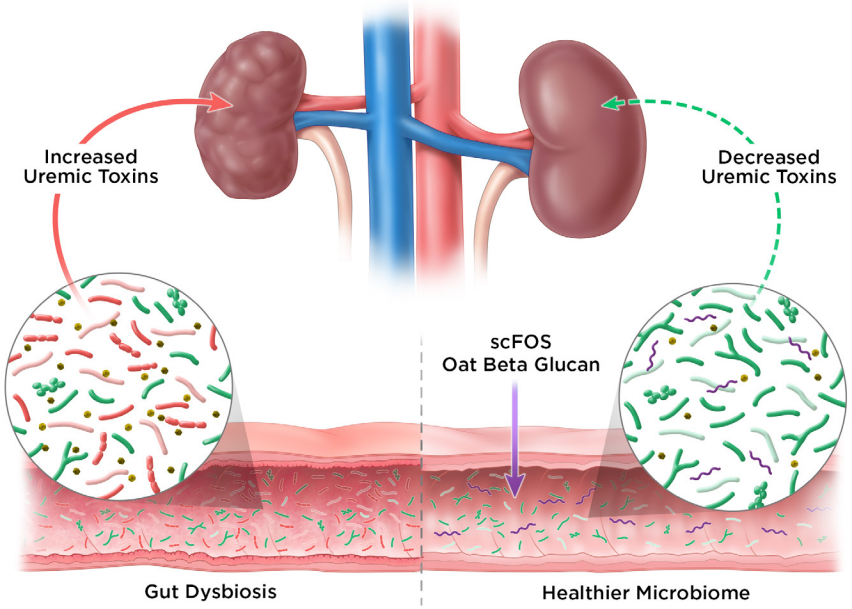


**Figure 2.** In chronic kidney disease (CKD), the microbiome balance shifts to proteolytic gut bacteria (red) that utilize dietary protein to produce uremic toxins (e.g., indoles, phenols, others), which can damage the kidneys (left side). Including appropriate amounts of prebiotic fibres (short-chain fructooligosaccharide or scFOS, and oat beta glucan) and betaine in a food for cats and dogs with CKD supports a shift to a healthier microbiome (right side), with increased abundance of saccharolytic bacteria (green) and decreased uremic toxins, which helps protect kidney health.

## *Feeding Controlled Amounts of High-Quality Protein Decreases Uremic Toxins*

In a study of cats with early CKD (IRIS Stage 1) who were fed similar amounts of phosphorus (0.5% as fed, 96 mg/100 kcal) and different amounts of protein for 111 days, those who consumed a food with lower amounts of protein had significantly decreased plasma concentrations of gut-derived uremic toxins **(Figure 3)**.[27] Indole sulfates including 3-indoxylsulfate, 5-hydroxyindole sulfate, 6-hydroxyindole sulfate, 3-hydroxyindolin-2-one sulfate and indoline-2-one increased with higher protein intake (P <0.0001). In addition, mean urine protein-to-creatinine ratio significantly decreased with lower dietary protein amounts.[27]

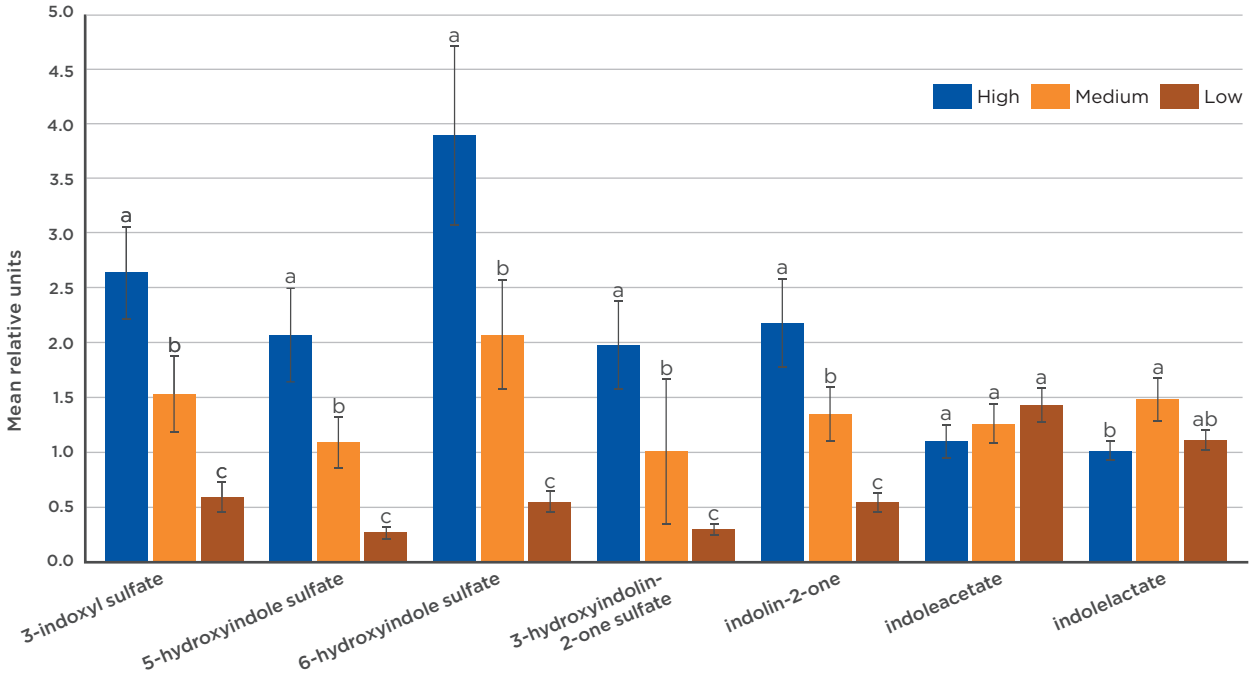


**Figure 3.** Relative mean units of plasma concentrations (and standard errors) of indoles after cats with CKD were fed higher protein (8.01 g/100 kcal), medium protein (7.01 g/100 kcal) and lower protein (5.33 g/100 kcal). *($P \leq 0.05$).*

## *Feeding Betaine and a Unique Prebiotic Fibre Blend Decreases Uremic Toxins and Provides Other Benefits*

Recent studies in cats and dogs with early CKD have demonstrated that including betaine and a unique blend of prebiotic fibres (ActivBiome+ Kidney Defense) (see box) results in beneficial effects including decreased uremic toxins, increased antioxidants, decreased markers of inflammation and oxidative stress, increased beneficial bacteria, and increased body mass.[9,10,28] Key findings from these studies are outlined below.

| **Hill's ActivBiome+ Kidney Defense** is a proprietary blend of betaine and prebiotics shown to activate the gut microbiome to help protect kidney function | | |
|---|---|---|
| **Ingredient** | **Site of Action** | **Description/Function** |
| Betaine | Kidney | A nutrient that functions as an osmolyte to support cell hydration and has antioxidant & anti-inflammatory properties |
| Oat beta glucan | Colon | Soluble, complex fibre that modulates the microbiome (colon) and helps reduce uremic toxins produced by gut bacteria |
| Short-chain fructooligosaccharides (scFOS) | Small intestine | Soluble, simple fibre that modulates the microbiome (small intestine) and helps reduce uremic toxins produced by gut bacteria |

## *Effects of Different Fibre Sources in Healthy Cats and Cats With CKD*

- A cross-over study was performed with 10 healthy cats and 10 cats with IRIS Stage 1 or 2 CKD.[9]
- Cats were fed dry Prescription Diet k/d Feline during a 2-week pre-trial period and then randomly assigned to two fibre treatments (pre-trial food with betaine, oat beta glucan and scFOS or pre-trial food with betaine, oat beta glucan and apple pomace) for 4 weeks each.
- At baseline, all cats with CKD had significantly higher circulating concentrations of creatinine, urea and gut-derived uremic toxins (e.g., indole sulfates) compared with healthy cats ($P \leq 0.05$).
- Cats with CKD had significantly lower concentrations of phenolic uremic toxins (guaiacol sulfate and 4-vinylphenol sulfate) after consuming the food with scFOS compared with the food containing apple pomace.
- Feeding scFOS increased beneficial bacteria in cats with CKD whereas apple pomace did not, suggesting that a more readily fermented fibre such as scFOS is preferred over apple pomace as a fibre source for cats with CKD.

## *A Therapeutic Renal Food With Added Betaine and Prebiotic Fibres Provides Benefits in Cats with CKD*

- Seven cats with CKD were fed Prescription Diet k/d for 4 weeks and then either k/d (control) or k/d with added betaine and prebiotic fibres (test) for 8 weeks before being crossed over to the other food for an additional 8 weeks.[28]
- There was no significant difference in food intake in cats with CKD (control vs test foods); however, total body mass (measured by DEXA) was significantly higher in cats with CKD after eating the test food compared with control food.
- Antioxidants (gamma-tocopherol and beta-tocopherol) were significantly increased after consumption of the test food.
- Uremic toxins produced by bacterial metabolism were significantly decreased in urine **(Table 1)** and increased in feces of cats with CKD when fed the test food.

**Table 1.** Relative concentrations of urine metabolites (known uremic toxins) that were significantly different ($P \leq 0.05$) in 7 cats with chronic kidney disease (CKD) after feeding control or test food for 8 weeks in a crossover study.[28]

| Urine Metabolites | Control Food (Prescription Diet k/d) | Test Food (Prescription Diet k/d with betaine and prebiotic fibres) | *P* Value |
|---|---|---|---|
| 5-hydroxyindole sulfate | 0.99 | 0.62 | 0.027 |
| 6-hydroxyindole sulfate | 1.06 | 0.64 | 0.037 |
| 7-hydroxyindole sulfate | 1.14 | 0.73 | 0.023 |

## *A Therapeutic Renal Food With Added Betaine and Prebiotic Fibres Provides Benefits in Dogs with CKD*

- 56 beagles (28 healthy, 28 with IRIS Stage 1 CKD) were fed a control food (Prescription Diet k/d) for 4 weeks and then randomly assigned to varying sequences of control food, a test food with low soluble fibre + betaine (Prescription Diet k/d with ActivBiome+ Kidney Defense) or a test food with high soluble fibre + betaine each for 10 weeks.[10]
- Dogs with CKD had significantly higher concentrations of creatinine, urea, indoles, advanced glycation end products and other gut-derived uremic toxins compared with healthy dogs. **(Figure 4)**.
- Compared with the control food, there was a significant decrease in uremic toxins after feeding both test foods to dogs with CKD ($P \leq 0.05$).
- In dogs with CKD, both test foods resulted in significant increases in antioxidant and polyunsaturated fatty acid markers and decreases in serum creatinine and an inflammatory marker compared with the control food.
- The abundance of the beneficial bacteria (phylum Bacteroidetes) was highest in dogs with CKD when fed the low soluble fibre test food.

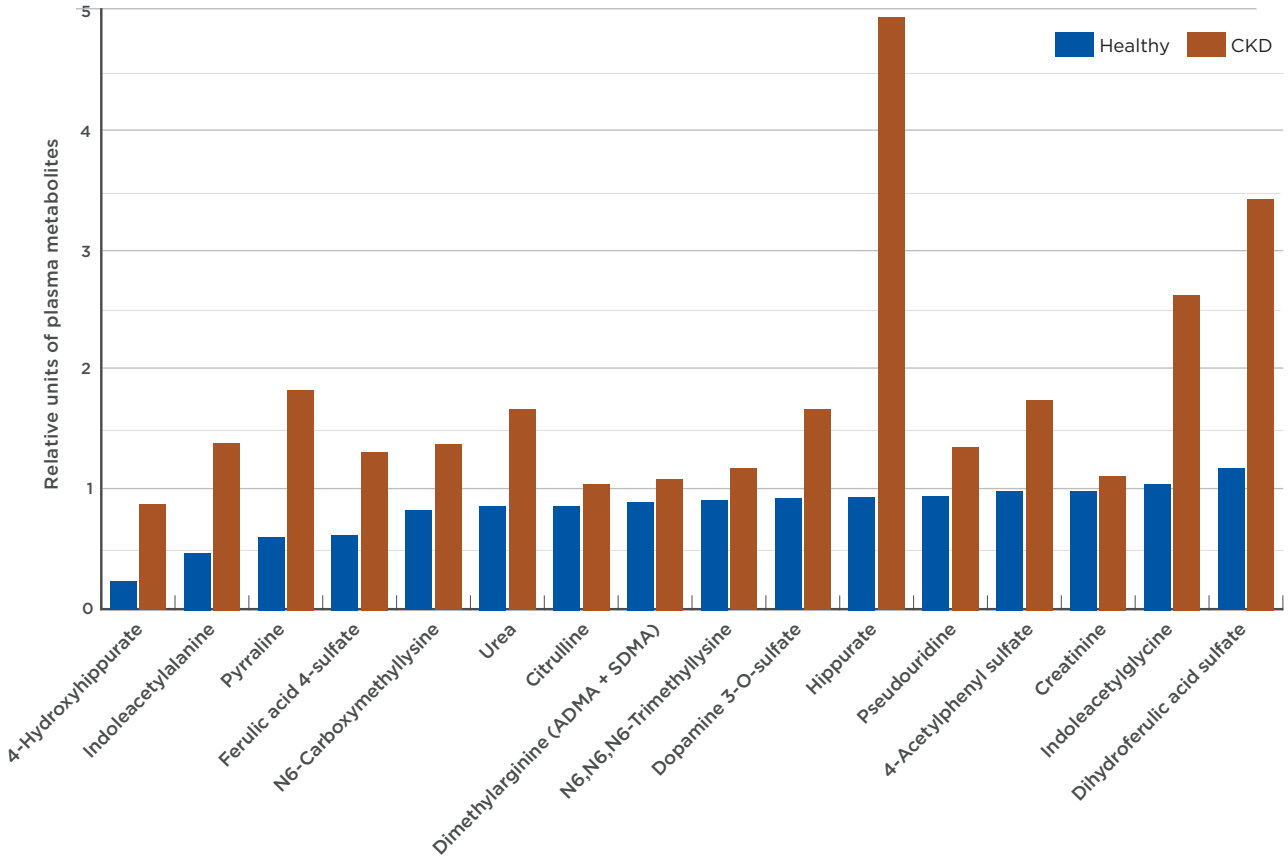


**Figure 4.** Dogs with early CKD (Stage 1) had significantly increased amounts of these gut-derived uremic toxins compared with healthy dogs ($P \leq 0.05$).

## Summary

Studies have shown that the microbiome of cats and dogs with CKD is significantly different from their healthy counterparts. This dysbiosis is characterized by a shift towards proteolytic metabolism by the microbiome, resulting in uremic toxin production, intestinal barrier disruption and further kidney injury. Nutritional interventions such as feeding decreased amounts of high-quality protein and adding betaine and a unique blend of prebiotic fibres (scFOS) to the **Hill's Prescription Diet k/d** (**Hill's Prescription Diet k/d ActivBiome+ Kidney Defense**) work with the microbiome of the pet to increase antioxidants, beneficial gut bacteria and body mass, while decreasing markers of inflammation, oxidative stress and uremic toxin concentrations.

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