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# Epakitin and Azodyl Clinical Studies

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# Making clinical sense

## CKD AND SECONDARY HYPERPARATHYROIDISM



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**Hyperphosphatemia and subsequent renal secondary hyperparathyroidism are recognized as serious complications of chronic kidney disease (CKD). Management of the rise in phosphorus can dramatically suppress disease progression. Studies in cats have shown that control of PTH extended life by almost 3 fold.<sup>1</sup>**

### PROGRESSION OF CKD

An important factor in the progression of CKD, the rise of parathyroid hormone (PTH) is seen as a trade-off for maintenance of initial (albeit transient) hyperphosphatemia and hypocalcemia (see **CKD & Hyperphosphatemia** on page 3).<sup>2,3</sup> Eventual exhaustion of greater than 75% to 80% of the functional nephron population leads to sustained hyperphosphatemia, direct stimulation of PTH synthesis and secretion, and altered vitamin D metabolism. PTH is also directly damaging to the kidneys.<sup>4</sup>

### THE ROLE OF VITAMIN D AND CALCITRIOL

Reduced nephron population mass contributes to decreased production of vitamin D to an active form, calcitriol (1,25-dihydroxycholecalciferol D3). Lack of calcitriol leads to parathyroid hyperplasia, nodular formation, and sequential aggressive synthesis and secretion of PTH. Reduced calcitriol also leads to reduced gut absorption and kidney reabsorption of calcium, contributing to lowered ionized calcium concentration, PTH elevation, and graduated hyperparathyroidism. Increased phosphorus binds remaining calcium, further driving down ionized calcium.<sup>5</sup>

### STAGING AND MANAGEMENT OF SECONDARY HYPERPARATHYROIDISM

CKD may be staged to facilitate appropriate therapy and monitoring using the IRIS CKD Staging System on page 3. Initial staging is based on fasting plasma creatinine and substages are based on accelerated plasma creatinine levels, along with serum phosphorus, protein spillage, and systemic blood pressure values. Along with application of low-phosphate diets and phosphate binders (our group initiates Epakitin® at IRIS Stage I), several management strategies are available for maintaining proper blood chemistry (see chart on page 4).<sup>6,7,8</sup> Except for advanced renal osteodystrophy (see Case Study on page 3), therapeutic dietary and medical management can often be rewarding in correcting the consequences of secondary hyperparathyroidism. When medical management fails, subtotal parathyroidectomy may be indicated.<sup>9</sup>

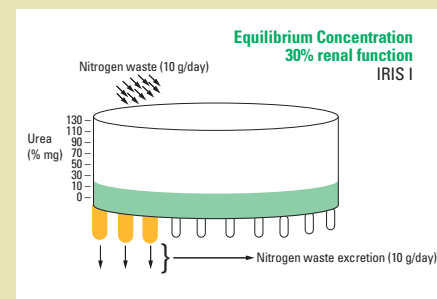
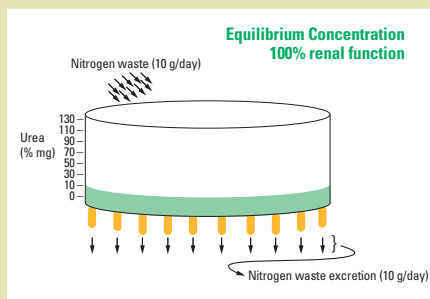
### Consequences of renal secondary hyperparathyroidism

- Anemia
- Metabolic acidosis
- Pruritus
- Hypertension
- Cardiac/vascular calcification
- Dystrophic mineralization
- Neurologic dysfunction
- Renal osteodystrophy

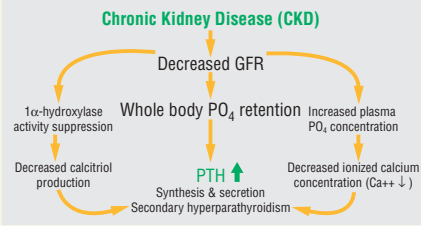
### Intact nephron hypothesis

The kidneys eliminate metabolic wastes from the body and maintain electrolyte and acid–base balance, blood pressure regulation, and hormone synthesis. The nephron is the functional unit. Although the intact nephron hypothesis states that each nephron is either a fully functional unit or does not function, surviving nephrons can undergo hypertrophy to increase their functional capacity. When nephrons are destroyed as in progressive CKD, the kidney's capacity to accommodate diminishes over time.

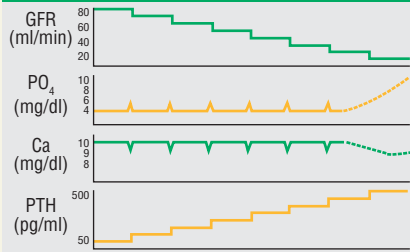
■ = Functional nephrons    ○ = Nonfunctional nephrons



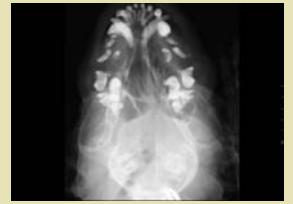
## CKD & Hyperphosphatemia (PO<sub>4</sub>↑)



## Progression of CKD Relationship of Ca, PO<sub>4</sub> & PTH



## Case study – Renal osteodystrophy



### Murphy

West Highland white terrier  
Male, 15 years old

### Results

The low ionized calcium and extremely high PTH are classic indicators of secondary hyperparathyroidism. Both radiographs show decalcification of the jaw ("rubber jaw"). The most common cause would be CKD (late IRIS III).

Laboratory indicator	Value	Range
Intact parathyroid hormone	> 210.00 pmol/L	3.00 – 17.00 pmol/L
Ionized calcium	1.16 mmol/L	1.25 – 1.45 mmol/L
BUN	93 mg/dL	< 20 mg/dL
Creatinine	4.8 mg/dL	< 1.2 mg/dL
SpGr	1.008	1.007 – 1.047

BUN = Blood urea nitrogen  
SpGr = Specific gravity

## IRIS\* CKD Staging System\*\*

Stage	Remaining renal function	Canine plasma creatinine (mg/dL)	Feline plasma creatinine (mg/dL)	Comments†
I	30%	< 1.4	< 1.6	<ul style="list-style-type: none"> <li>• Hyperphosphatemia unlikely</li> <li>• Hypertension possible</li> <li>• Proteinuria possible</li> </ul>
II	20%	1.4 – 2.0	1.6 – 2.8	<ul style="list-style-type: none"> <li>• Hyperphosphatemia possible</li> <li>• Hypertension possible</li> <li>• Proteinuria possible</li> </ul>
III	10%	2.1 – 5.0	2.9 – 5.0	<ul style="list-style-type: none"> <li>• Hyperphosphatemia probable</li> <li>• Hypertension possible</li> <li>• Proteinuria possible</li> </ul>
IV	< 5%	> 5.0	> 5.0	<ul style="list-style-type: none"> <li>• Hyperphosphatemia probable</li> <li>• Hypertension possible</li> <li>• Proteinuria possible</li> </ul>

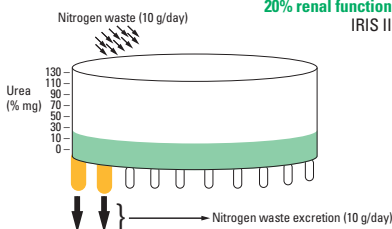
\* The mission of the International Renal Interest Society (IRIS) is to help veterinary practitioners better diagnose, understand, and treat renal disease in dogs and cats. IRIS and the IRIS Staging System were created at the 8th Annual Congress of the European Society of Veterinary Internal Medicine in Vienna, Austria, in 1998. Visit [iris-kidney.com](http://iris-kidney.com) for more information.

\*\* CKD can be further classified by presence or absence of proteinuria (P or Non-P) and presence or absence of systemic hypertension (H or Non-H); see comments column.

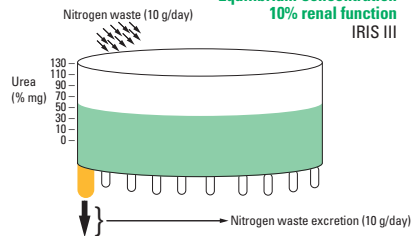
† Renal proteinuria defined as urine protein:creatinine ratio (dog > 0.5, cat > 0.4)

*For the most part, consequences of secondary hyperparathyroidism are associated with uncontrolled and sustained PTH elevation along with hyperphosphatemia.*

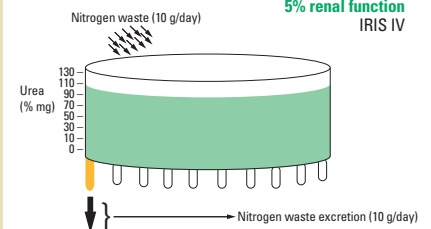
### Equilibrium Concentration 20% renal function IRIS II



### Equilibrium Concentration 10% renal function IRIS III



### Equilibrium Concentration 5% renal function IRIS IV



## Management of renal secondary hyperparathyroidism

### Reduce phosphorus in food

Several therapeutic foods are available that provide reduced levels of phosphorus to help slow the progression of kidney disease. They also have reduced levels of protein and may have some buffering capacity.

### Bind phosphate

Agent	Mode of action	Dose	Comments
Nutraceutical - Epakitin®	• Contains chitosan, calcium carbonate, lactose, and soy protein • Binds PO <sub>4</sub>	1 gm/5 kg Q 12 H with meals	• Begin using in IRIS Stage I • May need to be paired with additional PO <sub>4</sub> binders in IRIS Stages III and IV
Aluminum hydroxide	• Forms insoluble aluminum phosphate • Reduces PO <sub>4</sub> absorption	40-45 mg/kg Q 12 H with meals	• May be toxic <sup>10</sup> • May cause constipation, hypochromic microcytic anemia, myopathy, osteomalacia, progressive dementia
Calcium salts - Calcium carbonate - Calcium acetate	• Bind PO <sub>4</sub> in alkaline pH of small bowel	25-50 mg/kg elemental calcium Q 12 H with meals	• May cause hypercalcemia, soft tissue calcification, cardiovascular mineralization; restrict availability of calcitriol • Large volume demand as GFR decreases
Sevelamar HCl	• Noncalcium, nonaluminum PO <sub>4</sub> binder • Anion exchange resin trades Cl ions for PO <sub>4</sub> ions in small intestine	25-40 mg/kg Q 12 H with meals	• Potential to bind bicarbonate and cause metabolic acidosis • Potential to bind vitamins and bile acids
Lanthanum carbonate	• Noncalcium, nonaluminum PO <sub>4</sub> binder • Rare earth element not absorbed well from GI tract	40-50 mg/kg Q 12 H with meals	• GI disturbance • Cost

### Upregulate vitamin D3\*†

Cholecalciferol (vitamin D3)	• Provides substrate stores for 25-hydroxycholecalciferol	0.03 mg/kg/d for 2 days 0.02 mg/kg/d for 2 days then 0.01mg/kg/d maintenance	• Requires sequential liver and renal metabolism
Calcitriol	• Inhibits synthesis and secretion of PTH • Regulates calcium absorption from gut and reabsorption from kidney	1.5-3.5 ng/kg/d orally	• Hypercalcemia • Soft tissue calcification

### Partial parathyroidectomy

For resistant secondary hyperparathyroidism, removal of 3/4 of hyperplastic parathyroid gland and continuation of PO<sub>4</sub> binder therapy may be indicated.

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\* Optimum dosage must be determined based on serial evaluation of PTH activities and serum Ca and PO<sub>4</sub> levels. Paricalcitol and doxercalciferol are similar to calcitriol and currently being used in human medicine.

† Calcimimetic agents such as Sensipar® (Amgen) are being evaluated in treatment of secondary hyperparathyroidism as add-on therapy to calcitriol and vitamin D analogs and dietary phosphate binders.

### Epakitin® Highlights

By limiting the increase of phosphorus with the help of a low phosphorus diet and/or by adding a phosphate binder to the diet,<sup>1</sup> it is possible to slow down CKD and increase life expectancy. Contains a chitosan-based nutritional supplement (chitosan is a natural polysaccharide extracted from crab and shrimp shells).

- Binds phosphate
- Reduces uremia
- Improves general health of animals with CKD
- Demonstrated clinical efficacy



*This summary is based on a presentation at the 2009 NAVC Conference and references cited. The opinions expressed do not necessarily reflect the view of the publisher or the sponsor.*

# Effects of an Intestinal Phosphorus Binder on Serum Phosphorus and Parathyroid Hormone Concentration in Cats With Reduced Renal Function

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**KEY WORDS:** feline, kidney, phosphorus, nutrition

## ABSTRACT

Ten young adult cats, 6 intact males and 4 intact females, with reduced renal mass were randomized to receive either normal feline maintenance diet alone or diet plus phosphorus binder in a crossover design with each treatment period of 56 days in duration (Dietary Trial A). For Dietary Trial B, following a 3-month withdrawal period, 6 cats were placed on the intestinal phosphorus binder for 9 months to determine the long-term effects of this approach. During both Dietary Trials A and B, cats were studied by serum and urine biochemical testing and renal clearance studies. In Dietary Trial A, compared to diet alone, serum phosphorus and plasma parathyroid hormone concentrations were significantly lower when intestinal phosphorus binder was provided. There were no significant differences in blood urea nitrogen, serum creatinine concentration, glomerular filtration rate, or renal plasma flow. In Dietary Trial B, serum phosphorus and plasma parathyroid hormone concentrations were reduced at the 6- and 9-month

time points of the treatment period compared to measurements obtained before intestinal phosphorus binder administration. We conclude that the addition of an intestinal phosphorus binder to a normal maintenance feline diet can lower serum phosphorus and parathyroid hormone concentrations in cats with International Renal Interest Society stages I and II chronic kidney disease. The effect of the binder to reduce serum phosphorus concentration was present by Day 56 in Dietary Trial A and persisted at 6 and 9 months in Dietary Trial B.

## INTRODUCTION

Phosphorus retention and renal secondary hyperparathyroidism are common complications of feline chronic kidney disease (CKD).<sup>1-3</sup> Hyperphosphatemia has been associated with the development of renal structural lesions and renal secondary hyperparathyroidism in laboratory studies in rats,<sup>4</sup> dogs,<sup>5</sup> and cats<sup>6</sup> with induced kidney disease. In cats with spontaneous CKD, hyperphosphatemia is a negative prognostic factor.<sup>7,8</sup> Laboratory<sup>6</sup> and clinical<sup>1,2</sup> studies in cats have identified a renoprotective effect of dietary phosphorus restriction.

In a previous study, aged cats with spontaneous CKD were fed a maintenance feline ration combined with an intestinal phosphorus binder.<sup>9</sup> Results indicated that the binding agent was associated with reduced intestinal availability of phosphorus and reductions in plasma urea and phosphate concentrations. However, control animals were normal cats and it was difficult to separate time and treatment effects in this study. We hypothesized that hyperphosphatemia and renal secondary hyperparathyroidism could be managed in cats with early CKD through the addition of an intestinal phosphorus binding agent to a maintenance feline ration.

## **METHODS**

### **Animals**

Ten 8- to 11-month-old cats (4 intact females, 6 intact males) initially weighing  $3.64 \pm 0.20$  kg were procured from a commercial supplier (Liberty Research Inc., Waverly, New York, USA). Each cat's health was assessed by physical examination and determinations of plasma concentrations of blood urea nitrogen (BUN), serum creatinine (SCr), and the urine protein-to-creatinine ratio.

### **Animal Preparation**

All cats underwent right nephrectomy and infarction of approximately 5/6 of the left kidney by ligation of a variable number of branches of the renal artery, a procedure hereafter referred to as 11/12 nephrectomy,<sup>6,10</sup> approximately 6 months prior to the dietary trials.

### **Diets**

The cats were fed a canned feline maintenance diet (Purina Pro Plan Chicken and Liver Adult Cat Entrée, Nestle Purina PetCare Co., St Louis, Missouri, USA) throughout both Dietary Trials of the study and this diet contained approximately 0.47% phosphorus, 0.50% calcium, and 76% moisture on an as-fed basis. The cats were offered a pre-weighed amount of food daily, initially 35 kcal/kg body weight twice daily, with food being provided between 0800-1000h and between 1500-1700h daily. Daily

food intake was determined and amount of food provided was adjusted on a monthly basis with a goal of maintaining a stable body weight.

### **Phosphorus Binder**

A commercially available intestinal phosphorus binder containing chitosan and calcium carbonate (Epakitin, Vetoquinol, Buena, New Jersey, USA) was utilized in this study. The binder was mixed with food and provided twice daily at a dosage of 1 g for each meal for cats weighing <5 kg and 2 g twice daily for cats weighing >5 kg.

### **Dietary Trial A**

Approximately 6 months after renal mass reduction, the 10 cats were randomly divided into 2 groups of 5 each. For Days 1-56, cats in Group 1 received diet alone and cats in Group 2 received diet plus phosphorus binder. For Days 57-112, the groups were crossed over so that Group 1 received diet plus phosphorus binder and Group 2 received diet alone. Biochemical measurements were obtained prior to 11/12 nephrectomy (pre), at the time of division into treatment groups before drug administration (Day 0), and at Days 26, 56, 84, and 112. Glomerular filtration rate (GFR), renal plasma flow, and fractional excretion of phosphorus measurements were obtained at Days 0, 56, and 112. Serum parathyroid hormone concentration (PTH) was determined prior to 11/12 nephrectomy (pre) and on Days 56 and 112.

### **Dietary Trial B**

Six cats (2 males and 4 females) with International Renal Interest Society (IRIS) stages I or II CKD were selected from the original group of 10 for inclusion in a second dietary trial of 15 months' duration. For this trial, the cats received diet (Purina Pro Plan Chicken and Liver Adult Cat Entrée, Nestle Purina PetCare Co., St Louis, Missouri, USA) only for 3 months (time -3 to 0 months), diet plus binder for 9 months, and subsequently diet only for 3 months. Immediately prior to institution of binder administration (Month 0) and at 3, 6, 9, and

12 months later, SCr, serum phosphorus, serum calcium, BUN, PTH, GFR, and urine protein-to-creatinine ratio measurements were obtained.

### Biochemical Measurements

Blood was obtained by venipuncture for subsequent measurement of serum concentrations of phosphorus, calcium, BUN, and SCr. Serum analyte determinations were obtained by a semi-automated device (Spectrum CCX, Abbott Diagnostics, Irving, Texas, USA).

### Renal Clearance Studies

The GFR and renal plasma flow were estimated as urinary clearance of exogenously administered creatinine and para-aminohippuric acid, respectively, utilizing a previously described procedure.<sup>10</sup> During this procedure, urine was obtained by urethral catheterization for urinalysis and determination of the urine protein-to-creatinine ratio. Serum and urine creatinine and urine protein determinations were obtained by a semi-automated device (Spectrum CCX, Abbott Diagnostics, Irving, Texas, USA). Serum and urine concentrations of para-aminohippuric acid was determined by a chemical method.<sup>10</sup> Fractional clearance of phosphorus (%) was determined during these procedures as  $100 \times \text{urinary phosphorus excretion (mg/min)}/\text{GFR (mL/min)} \times \text{serum phosphorus concentration (mg/mL)} / \text{filtrate phosphorus content}$ .

### Statistical Analysis

Values are reported as mean  $\pm$  SEM. Statistical analyses were performed with the aid of a commercial software package (Statview 4.5, Abacus Concepts, Inc., Berkeley, California, USA). Statistical comparisons were by analysis of variance with inclusion of an effect for drug treatment and time (if multiple measurements were made) in the model. If a statistically significant global effect was observed, pairs of group means were compared by Fisher's protected least significant different test. A *P* value of  $<0.05$  was taken as indicative of a statistically significant difference.

## RESULTS

### Dietary Trial A

The mean body weight for the 10 cats was  $3.85 \pm 0.28$  kg on Day 0, prior to treatment. The average quantity of food offered was  $120.5 \pm 8.3$  g per cat and there were no significant treatment or time effects in quantity of food offered, food intake, or body weight (Table 1). The initial mean values for parameters (Day 0) for cats in this trial were  $3.36 \pm 0.27$  g albumin/dL,  $35.1 \pm 3.6$  IU alanine transferase/L,  $154 \pm 2$  mEq sodium/L,  $3.6 \pm 0.1$  mEq potassium/L,  $18.0 \pm 0.8$  mmol bicarbonate/L,  $28.0 \pm 3.3$  IU alkaline phosphatase/L, and  $2.1 \pm 0.1$  mg magnesium/dL. The hematocrit averaged  $27.2\% \pm 1.2\%$  and the urine specific gravity was  $1.023 \pm 0.003$ . There was no significant time or treatment effect for these serum and urine parameters during Dietary Trial A.

On Day 0, GFR was  $1.42 \pm 0.11$  mL/min/kg body weight and renal plasma flow was  $5.00 \pm 0.34$  mL/min/kg. Analysis of serum of the 10 cats revealed mean values of  $5.85 \pm 0.22$  mg phosphorus/dL,  $10.4 \pm 0.1$  mg calcium/dL,  $2.56 \pm 0.12$  mg creatinine/dL, and  $45.5 \pm 2.7$  mg BUN/dL on Day 0. There were no significant treatment or time effects on SCr, BUN, GFR, or renal plasma flow. There was a significant ( $P < 0.05$ ) treatment effect on serum phosphorus concentration and urinary fractional clearance of phosphorus during the trial with lower values for both observed during administration of the intestinal phosphorus binding agent (Table 1). Compared to diet alone, provision of the intestinal phosphorus binder was associated with a significantly ( $P < 0.05$ ) lower PTH of approximately 46% overall. However, the mean PTH in cats with reduced renal mass fed the binder remained significantly ( $P < 0.05$ ) greater than results of measurements obtained in the cats prior to renal mass reduction ( $2.9 \pm 0.5$  pmol/L).

### Dietary Trial B

Following a 3-month washout period, the cats were provided with a mean phosphorus binder intake of  $0.21 \pm 0.02$  g/kg body

weight twice daily for 9 months (Months 0-9). The cats then received diet alone for 3 months. There was no significant treatment or time effect on food intake or body weight. Compared to Month 0, the mean values for serum phosphorus were lower ( $P < 0.05$ ) at 6 and 9 months (Table 2). While 4 of 6 cats had serum phosphorus concentrations between 4.5 mg/dL and 5.1 mg/dL after 3 months of administration of the intestinal phosphorus binding agent, all serum phosphorus concentrations were between 3.0 and 4.5 mg/dL for Months 6 and 9. There were no significant treatment or time effects on other serum biochemical parameters, GFR, or the urine protein to-creatinine ratio.

In a previous laboratory study utilizing the remnant kidney model of CKD in cats, ingestion of a phosphorus supplemented diet exacerbated hyperphosphatemia and hyperparathyroidism and was associated with more severe renal structural lesions.<sup>6</sup>

A more recent study of spontaneous feline CKD demonstrated that elevated serum phosphorus concentration was associated with a shorter survival time in cats with CKD.<sup>8</sup> Dietary phosphorus restriction coupled with the use of an aluminum-based intestinal phosphorus binding agent reduced serum phosphorus, reduced plasma PTH concentration, and increased median survival time.<sup>2</sup> As a consequence of these and

**Table 1:** Results at the end of a 56-day treatment period of measurements of parameters in cats with reduced renal mass fed diet alone or diet plus intestinal phosphorus binder in Dietary Trial A.

Parameter	Diet Alone	Diet Plus Binder
Number of cats	10	10
Food intake (g/kg body weight)	106 ± 7	107 ± 7
Phosphorus binder intake (g/kg body weight, twice daily with food)	0	0.23 ± 0.02
Body weight (kg)	3.83 ± 0.26	3.84 ± 0.24
GFR (mL/min/kg body weight)	1.49 ± 0.11	1.41 ± 0.09
Renal plasma flow (mL/min/kg body weight)	4.88 ± 0.78	4.69 ± 0.56
SCr (mg/dL)	2.54 ± 0.05	2.68 ± 0.07
BUN (mg/dL)	42.7 ± 1.2	43.5 ± 1.7
Serum phosphorus (mg/dL)	5.55 ± 0.11	5.14 ± 0.11*
Serum calcium (mg/dL)	10.4 ± 0.1	10.4 ± 0.1
Serum parathyroid hormone (pmol/L)	15.3 ± 1.6	8.3 ± 1.2*
Urinary fractional clearance of phosphorus (%)	38.6 ± 4.1	27.9 ± 2.9*
Urine protein/creatinine ratio	0.42 ± 0.05	0.42 ± 0.08

GFR = glomerular filtration rate; SCr = serum creatinine; BUN = blood urea nitrogen.

\* $P < 0.05$  vs corresponding mean value for diet alone.

## DISCUSSION

Our study demonstrated an effect of an intestinal phosphorus binder to lower serum phosphorus concentration, PTH, and urinary fractional excretion of phosphorus in cats fed a maintenance feline diet. This effect was observed in both a 56-day crossover trial and during a 9-month trial in a subset of these cats. The cats were fed a maintenance diet with phosphorus content higher than typically recommended<sup>11</sup> for azotemic cats.

other studies of phosphorus homeostasis in CKD, dietary phosphorus restriction is routinely recommended for cats with azotemic CKD corresponding to IRIS stages II-IV.<sup>11</sup>

A previous study<sup>9</sup> of the same intestinal phosphorus binding agent in cats, which included 6 older animals with spontaneous azotemia, demonstrated a reduction in apparent digestibility of phosphorus. In that study, both BUN and plasma phosphorus concentration decreased when the cats were provided the intestinal phosphorus binder.

**Table 2:** Results of measurements of parameters in cats with reduced renal mass fed diet plus intestinal phosphorus binder for Months 0-9 and diet alone for Months 10-12 in Dietary Trial B. The cats received diet alone for 3 months prior to the initiation of this trial.

Parameter	0 Months	3 Months	6 Months	9 Months	12 Months
Number of cats	6	6	6	6	6
Body weight (kg)	3.88 ± 0.34	3.85 ± 0.30	3.88 ± 0.26	3.90 ± 0.30	3.85 ± 0.29
GFR (mL/min/kg)	1.40 ± 0.15	1.36 ± 0.08	1.35 ± 0.07	1.37 ± 0.11	1.34 ± 0.13
SCr (mg/dL)	2.50 ± 0.16	2.68 ± 0.16	2.77 ± 0.14	2.50 ± 0.16	2.62 ± 0.15
BUN (mg/dL)	38.2 ± 3.2	43.2 ± 4.2	35.0 ± 3.3	35.8 ± 3.1	41.8 ± 2.4
Serum calcium (mg/dL)	10.9 ± 0.1	10.9 ± 0.2	10.7 ± 0.1	10.9 ± 0.1	10.5 ± 0.1
Serum phosphorus (mg/dL)	5.0 ± 0.22	4.62 ± 0.31	3.52 ± 0.26*	3.87 ± 0.23*	4.73 ± 0.19
Serum parathyroid hormone concentration (pmol/L)	12.7 ± 2.9	7.9 ± 1.4	5.3 ± 0.9*	4.9 ± 1.2*	9.0 ± 1.2
Urine protein/creatinine ratio	0.33 ± 0.06	0.39 ± 0.06	0.41 ± 0.03	0.35 ± 0.05	0.34 ± 0.04

GFR = glomerular filtration rate; SCr = serum creatinine; BUN = blood urea nitrogen.

\* $P < 0.05$  vs corresponding value for 0 months.

As a feline maintenance diet was also used in this previous study, the authors suggested that this approach could be an alternative treatment option for cats refusing to ingest diets specially formulated for CKD. While our study demonstrated a decrease in serum phosphorus concentration with a similar dietary approach, there was no apparent effect of the binding agent on BUN, SCr, GFR, renal plasma flow, or proteinuria. We did not establish the long-term effects of the use of a maintenance diet plus intestinal phosphorus binder on the preservation of renal structure and function in cats. Thus, we are not advocating the use of the present dietary approach as an alternative to specially formulated diets. It is important to note that studies<sup>1,12</sup> of cats with spontaneous IRIS stages II and III CKD demonstrated a beneficial effect of dietary modification, specifically a decrease in uremic episodes, renal-related deaths, or prolonged survival. The beneficial effects observed in these studies may have been due to modified phosphorus content to other dietary variables present. Nonetheless, cats with CKD may find specially formulated diets to be less acceptable or owners may offer a mixture of diets and if so, our study confirmed previous results<sup>9</sup> that a reasonable degree of control of phosphorus metabolism can be achieved with the addi-

tion of an intestinal phosphorus binder to a feline maintenance diet, particularly in IRIS stages I and II.

Our study demonstrated that cats with azotemia equivalent to IRIS stages I and II CKD and very mild changes in serum phosphorus concentration had serum phosphorus concentrations lowered after the addition of an intestinal phosphorus binding agent while being fed a normal feline maintenance diet. It may be beneficial to lower serum phosphorus concentration below the upper level of the normal range in dogs and cats with CKD. The goal for control of serum phosphorus concentration in CKD has been proposed to be as low as 4.5 mg/dL.<sup>11</sup> At 6 and 9 months of administration during Dietary Trial B, we achieved this level of control in the cats with induced azotemia comparable to IRIS stages I and II. Cats with more severe azotemia, comparable to IRIS stages III and IV, are unlikely to achieve this level of control of serum phosphorus concentration with the present dietary approach.

Hyperparathyroidism is present in CKD and has been linked to uremia or disease progression in various studies in other species.<sup>13-15</sup> In the present study, the addition of the intestinal phosphorus binding agent to a canned maintenance feline diet reduced the magnitude of hyperparathyroidism.

However, compared to values obtained in the same cats on the same diet prior to renal mass reduction, the PTH remained elevated. Control of serum phosphorus concentration and prolongation of survival in cats with CKD has previously been associated with reduction of PTH.<sup>2</sup>

The addition of an intestinal phosphorus binding agent to food may reduce food intake in azotemic cats. In the present study, this particular agent containing calcium carbonate and chitosan had no such effect. In summary, the addition of an intestinal phosphorus binding agent to a canned feline maintenance diet reduced serum phosphorus and PTH in cats with induced azotemia equivalent to IRIS stages I and II.

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## Effects of a dietary chitosan and calcium supplement on Ca and P metabolism in cats

### *Effekte der Zufütterung einer Chitosan- und Kalziumkombination auf den Kalzium- und Phosphorstoffwechsel der Katze*

Elisabeth Wagner<sup>1</sup>, Ilse Schwendenwein<sup>2</sup>, Jürgen Zentek<sup>1</sup>

#### Summary

The aim of the present study in cats was to investigate the potential effects of a calcium carbonate and chitosan supplement on blood parameters in aged cats with moderate chronic renal failure and on the mineral balance in adult healthy cats. For the trials, 10 neutered cats 2–4 years of age were fed for 21 days and six neutered cats (2 male and 4 female), 14 years of age, with elevated urea and phosphorus level in the plasma were fed for 35 days with a supplement. The apparent digestibility of phosphorus was ( $p < 0.05$ ) reduced in the treatment period. Plasma urea inorganic phosphate decreased significantly ( $p < 0.05$ ) in the old cats after 35 days of treatment. The treatment had a significant effect on the phosphorus, gross energy, dry matter, crude ash, crude fiber and crude protein digestibility in adult healthy cats. The practical implication could be an alternative treatment option for cats refusing to ingest veterinary renal diets.

**Keywords:** chronic renal failure, chitosan, cat, plasma urea, plasma phosphate, phosphorus binder, digestibility

#### Zusammenfassung

Das Ziel der Studie war der Nachweis des Einflusses eines Supplements mit Kalziumkarbonat und Chitosan auf die Blutparameter bei Katzen mit erhöhten Harnstoff- und Phosphatgehalten im Plasma und auf den Mineralstoffhaushalt bei gesunden Katzen. Für die Studie wurden 10 kastrierte Katzen (2–4 Jahre alt) über 21 Tage und 6 kastrierte Katzen (14 Jahre alt) mit einem erhöhten Harnstoff- und Phosphorgehalt im Plasma 35 Tage mit einem entsprechenden Futterzusatz gefüttert. Dieser wurde einem kommerziellen Dosenfutter zugesetzt. Die scheinbare Verdaulichkeit des Phosphors war in der Supplementierungsphase gegenüber der Kontrollperiode reduziert ( $p < 0.05$ ). Die mittlere Harnstoffkonzentration und ebenso der anorganische Phosphorgehalt im Plasma wurde nach 35 Tagen mit dem Zusatz signifikant ( $p < 0.05$ ) abgesenkt. Es wurde ein signifikanter Einfluss dieses Supplements auf die Verdaulichkeit der Bruttoenergie, der Trockensubstanz, der Rohasche, der Rohfaser, des Rohproteins und des Phosphats bei gesunden Katzen nachgewiesen. Diese Supplementierung könnte für Katzen, die eine Nierendiät benötigen würden, eine weitere diätetische Alternative sein.

**Schlüsselwörter:** chronische Niereninsuffizienz, Chitosan, Katze, Plasmaharnstoff, Plasmaphosphat, Phosphorbinder, Nährstoffverdaulichkeit

## Introduction

Chronic renal failure (CRF) is the most common renal disease in dogs and cats. It is characterized by progressive and irreversible renal structural lesions and nephron losses. Patients with CRF show clinical and biochemical signs, depending on the progression of the disease. Laboratory findings are azotemia or uremia, including metabolic acidosis and hyperphosphatemia. Serum creatinine and blood urea nitrogen (BUN) concentrations are commonly used screening tests. Serum creatinine and urea levels are negatively correlated with the glomerular filtration rate.

Phosphorus is absorbed from the gastrointestinal tract and primarily excreted by the kidneys. Renal excretion reflects the net effect of glomerular filtration and tubular resorption. If dietary phosphorus intake remains constant, a decline in the glomerular filtration rate will lead to phosphorus retention and hyperphosphatemia and consequently to renal hyperparathyroidism. Hyperphosphatemia can be managed by restricted dietary phosphorus intake. If this is not sufficient to reduce increased serum inorganic phosphate levels, oral administration of intestinal phosphorus binding agents is recommended (Polzin et al. 1995). Traditionally, phosphate binders containing aluminium have been used effectively to control serum phosphorus levels in patients with chronic renal failure. Barber et al. (1999) and Elliott et al. (2000) combined a veterinary diet restricted in phosphorus and protein with or without an intestinal phosphate binding agent (aluminium hydroxide) and investigated plasma phosphate and parathyroid hormone (PTH) concentrations in cats. Phosphate binders reduced serum phosphorus and PTH levels. Several studies evaluated the phosphate binding capacity of calcium carbonate (Slatopolsky et al., 1986), aluminium hydroxide (Takamoto et al., 1985) and calcium acetate (Janssen et al., 1996; Lau et al., 1998) in humans with renal failure. Addition of ferrihydrite, calcium acetate (Weaver et al., 1999) and zirconyl chloride octahydrate (Graff and Burnel, 1995) was found to be effective in rats. High phosphate concentrations are also reduced by iron (III) hydroxide complex (Yamaguchi et al., 1999). Rats (15 male Wistar rats) were fed for 7 days with a rodent chow containing 0, 1, 4 and 8 % of the iron (III) hydroxide complex. The urinary phosphorus levels dropped significantly ( $p < 0.01$ ) in a dose dependant manner.

The effects of chitosan-coated dialdehyde cellulose as an oral adsorbant of urea and ammonia were examined in rats with adriamycin induced progressive chronic renal failure (CRF) (Nagano et al., 1995). Chitosan (5 %) or a charcoal adsorbent (5 %) were fed over four months. CRF rats fed the normal diet and the charcoal adsorbent developed progressive azotemia, hyperphosphatemia, proteinuria and anaemia. After 9 weeks increased number of losses occurred. In contrast, chitosan treated rats had decreased blood urea nitrogen, serum creatinine and serum phosphate and longer survival periods.

Furthermore Jing et al. (1997) investigated the effects of chitosan on eight haemodialysis patients from 30 to 72 years of age. After a pre period of 1 week the patients received 30 chitosan tablets (45 mg chitosan/tablet) three times a day. Significant reductions in urea and creatinine levels in serum were observed after 4 weeks of chitosan ingestion. After 12 weeks physical strength, appetite and sleep had been improved.

The effect of chitosan on Calcium ( $\text{Ca}^{47}$ ) metabolism was investigated in male Wistar rats by Wada et al. (1997). Rats were fed a 5 % chitosan diet for 40 days and whole body retention of  $\text{Ca}^{47}$  was significantly decreased compared with rats fed a cellulose diet, but showed no significant difference to rats fed a fibre-free diet. The urinary excretion of  $\text{Ca}^{47}$  was significantly increased in the chitosan group when compared with the cellulose group.

In rats fed an iron (III) chitosan complex serum phosphorus levels were significantly reduced after 15 days. The faecal phosphorus levels were higher ( $p < 0.01$ ), while urinary phosphorus was not significantly reduced (Baxter et al. 2000).

Studies in dogs and cats on the effects of calcium carbonate in combination with chitosan have not been published to our knowledge. In cats, practical problems occur frequently due to low palatability of veterinary diets in patients with chronic renal failure. Alternatives to the standard dietary treatment would be useful especially in cases with mild signs of renal failure.

The aim of the present study in cats was to investigate the potential effects of calcium carbonate and chitosan on blood parameters in aged cats with biochemical signs of chronic renal failure and on the calcium and phosphorus balance in adult healthy cats.

## Material and Methods

### Balance Trial

Animals: Ten cats (8 female and 2 male neutered) were allocated into single cages (1 × 1 m, 3 m height). The age of the cats ranged from two to four years. The cats did not show clinical signs of renal dysfunctions. Vaccinations and deworming procedures were performed according to established protocols.

Diet: A commercial canned diet (Whiskas senior; Masterfoods Austria OHGA-2460 Bruck/Leitha) was fed. Crude nutrients, gross energy, calcium and phosphorus content of the diet are summarized in table 1. The amount of food was adjusted to meet maintenance requirements of the cats.

In the treatment period chitosan and calcium carbonate (Ipakitine<sup>®</sup>, Vetoquinol, Lure Cedex, France; composition: 8 % crab shell extract; 10 % calcium carbonate and 82 % lactose; ingredients: crude ash 9.9 %, crude protein 4 %, ether extract 0.55 %, crude fiber <0.1 %, potassium 0.015 %, calcium 3.7 % and phosphorus <0.01 %) was added to the canned diet. The applied dosage was 1 g/5 kg body mass/twice daily. The control and the treatment period lasted 21 days.

Procedures: Urine was collected from day 7 to 14. The urine pH, volume and the calcium and phosphorus concentrations were measured. The 24 hour urine collection was done under a thin layer of thymol/mineral oil solution to prevent losses by microbial activities and evaporation.

A digestibility trial was performed from days 14 to 21.

Analytical methods: Proximate analysis in feed and faeces was performed according to the procedures of Naumann and Bassler (1993). Minerals were measured in faeces and urine after wet ashing of the samples in a mixture of a 40 %  $\text{HNO}_3$  (65 %), 10 %  $\text{HCl}$  (35 %), 20 %  $\text{H}_2\text{O}_2$  (30 %) and 30 % water in a microwave oven (MLS GmbH.; MLS-Ethos plus, Leutkirch im Allgäu, Ger-

**TABLE 1:** Crude nutrients, gross energy and minerals of the canned diet fed to the cats.

		fresh matter	dry matter
dry matter	%	17.2	100
crude ash	%	1.25	7.24
crude protein	%	8.27	48.0
ether extract	%	3.77	21.9
crude fiber	%	0.66	3.80
nitrogen free extract	%	3.28	19.0
gross-energy	MJ/kg	4.05	23.50
calcium	g/kg	2.97	17.2
phosphorus	g/kg	1.54	8.92

**TABLE 2:** Dry matter intake and weight of the faeces (g DM/d) of the experimental cats in the balance trial.

period		feed	faeces
control	mean	45.7	8.8
	SD	7.4	0.5
treatment	mean	43.8	11.5
	SD	7.7	0.5

**TABLE 3:** Apparent digestibility of crude nutrients during the treatment period (supplementation with calcium carbonate/chitosan) and the control period in the experimental cats in the balance trial.

period		dry matter		crude ash	crude protein	ether extract	crude fiber	nitrogen free extract	gross-energy
		%	%	%	%	%	%	%	%
control	mean	8.1	50.1	83.8	93.6	58.5	76.2	85.1	
	SD	5.6	21.4	5.0	3.2	17.0	7.4	7.0	
treatment	mean	74.7*	27.9*	79.2*	91.6	30.6*	70.5	78.1*	
	SD	5.0	14.9	4.8	1.6	19.6	8.1	4.4	

\* (p &lt; 0.05)

**TABLE 4:** Apparent digestibilities of phosphorus and calcium in the experimental cats in the balance trial

period		phosphorus	calcium
control	mean	38.1	21.9
	SD	17.8	22.1
treatment	mean	17.2*	- 4.5*
	SD	19.6	22.0

\* (p &lt; 0.05)

**TABLE 5:** Intake and urinary excretion of calcium and phosphorus in the experimental cats in the balance trial.

period		intake		excretion	
		calcium mg/kg	phosphorus BW/day	calcium mg/kg	phosphorus BW/day
control	mean	187.9	97.4	0.9	103.8
	SD	39.5	20.5	0.7	22.1
treatment	mean	197.8	93.4	0.8	87.6
	SD	39.9	19.9	0.6	15.3

many). Calcium was measured by atomic absorption spectrometry (Perkin Elmer 3030B, Wellesley, USA), phosphorus by the vanadate molybdate method using a spectrophotometer (Hitachi U 3000, Tokyo, Japan).

#### Effects of chitosan with calcium carbonate in older cats with increased plasma urea and phosphorus

**Animals:** Six cats, average age 14 years (2 male and 4 female neutered), with an increased urea and phosphorus concentration in the plasma. Vaccinations and deworming procedures were done regularly, the 6 cats were allocated in a common kennel in the Institute of Nutrition (12.3 m<sup>2</sup>, 3 m height).

**Diet:** Cats were fed with the diet as described in table 1 for at least 5 weeks before blood sampling. All cats were subsequently fed the identical diet supplemented with chitosan and calcium carbonate for 35 days before blood samples were taken (1 g/5 kg body mass/twice daily).

**Sampling:** Blood was taken from the Vena cephalica antebrachii into lithium-heparin vials (Greiner Bio one GmbH, Kremsmünster, Austria). Differential blood counts and biochemistry (urea, creatinin, total protein, calcium, phosphorus, aspartate aminotransferase, alanine amino-transferase) were done by the central laboratory of the University of Veterinary Medicine, Vienna.

**Analytical methods:** All biochemical tests were run on an automated Hitachi 911 analyzer (Hitachi, Tokyo, Japan): Urea (Urease, Roche Diagnostics 1 489 364), creatinin (Jaffé compensated kinetic, Roche Diagnostics 1 489 291), total protein (Biuret, Roche Diagnostics 1 553 836), calcium (Kresolphthalein-Komplexon, Roche Diagnostics 1 489 216), phosphorus (Molybdat, Roche Diagnostics 1 489 348), aspartate aminotransferase (AST, opt. Standardmethode DGKC, Roche Diagnostics 816 337), alanine aminotransferase (ALT, opt. Standardmethode DGKC, Roche Diagnostics 816 442). Differential blood counts were performed by laser flow cytometry (Advia 120™, Bayer Diagnostics, Wien) and, if necessary, by microscopic control.

#### Statistics

Data are expressed as means and standard deviations (SD). Group differences were evaluated by Student t-test (WinSTAT® Microsoft Excel® 1999) and in the old cats by a paired Student t-test (p < 0.05).

## Results

#### Balance trial

The calcium intake of the cats during the experimental period was 830 mg/day (SD = 132.3) and in the control period 787 mg/day (SD = 128.3). The mean phosphorus intake of the cats was 392 mg/day (SD = 68.6) and in the control period 408 mg/day (SD = 66.5). The mean dry matter intake, faeces weight are presented in table 2 and the apparent digestibilities of crude nutrients are presented in table 3. Dry matter, crude protein, crude fiber, crude ash and gross-energy digestibility decreased significantly (p < 0.05), but the variation between the individuals was high.

The apparent digestibility of phosphorus was significantly ( $p < 0.05$ ) reduced in the experimental period compared to the control period (table 4). The apparent digestibility of calcium was slightly negative in the treatment period and reached 21.9 % in the control period.

The urine volume during the treatment period was 130,2 ml/day (SD= 26,0) compared to 134,0 ml/day (SD = 27,5) in the control period. The pH was in both feeding periods around 7.8 (SD = 0.3). The urinary excretion of phosphorus and calcium was not significantly influenced by the dietary treatment (table 5).

**Effects of chitosan with calcium carbonate in older cats with increased plasma urea and phosphorus**

The mean urea concentration in the plasma of the old cats was 85.6 mg/dl (SD = 18.1) at day 1 (reference 20–65 mg/dl) and was ( $p < 0.05$ ) reduced to 61.2 mg/dl (SD = 11.4) after 35 days of treatment. Figure 1 demonstrates the urea concentrations for each cat at the beginning and after 35 days of the treatment. Plasma inorganic phosphate (figure 2) decreased significantly ( $p < 0.05$ ) after 35 days of treatment from 1.7 mmol/l (SD = 0.2) at day 1 to 1.1 mmol/l (SD = 0.3) at day 35 (normal range 0.8-1.6 mmol/l). Practically no change was observed in the plasma calcium content after 35 days of treatment. The mean calcium concentration was 2.8 mmol/l (SD = 0.2) at days 1 and 35. At the beginning and at the end of the treatment the plasma creatinine levels were comparable with a mean of 1.2 mg/dl (SD = 0.1). Total protein and packed cell volume were in the normal ranges. The experimental cats had an increased activity of alanineamino-transferase at the beginning (105.8 U/l, SD = 58.6). The results of the haematology and blood chemistry are summarized in tables 6 and 7.

**Discussion**

The aim of the study was to investigate the potential effects of a dietary supplement containing calcium carbonate and chitosan on blood parameters in aged cats with biochemical signs of moderate chronic renal failure and on the mineral balance in adult healthy cats. Cats are highly susceptible to develop chronic renal failure with increasing age. Dietetic treatment is mainly based on moderate protein restriction in combination with decreased phosphorus intake (Barber et al., 1999; Elliot et al., 2000; Polzin et al., 2000). Unfortunately, palatability problems

**TABLE 6:** Blood chemistry of the 6 older cats before (day 1) and after 35 days of treatment.

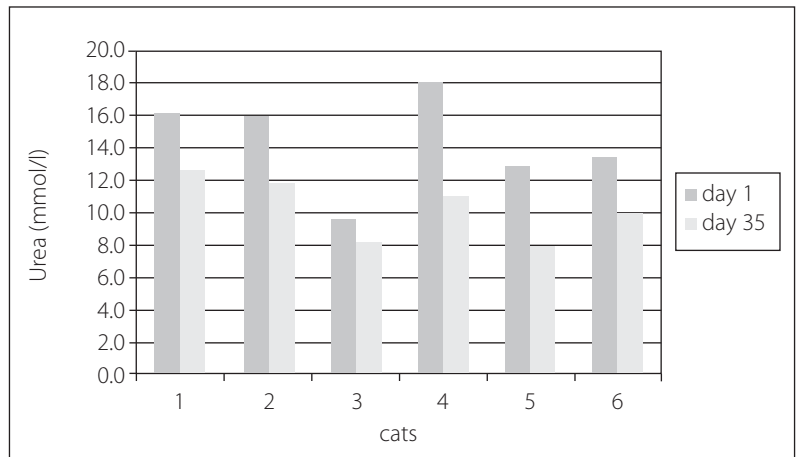
period		urea mmol/l	creatinine mmol/l	total proein g/l	AST U/l	ALT U/l	calcium mmol/l	phosphorus mmol/l
control	mean	14.3	106.1	84.6	38.8	105.8	2.8	1.7
	SD	3.0	11.2	7.85	43.5	58.6	0.16	0.17
treatment	mean	10.2*	112.0	87.7	18.7	103.5	2.8	1.1*
	SD	1.9	27.2	6.49	12.2	92.5	0.08	0.28

\*( $p < 0.05$ ); AST = aspartate aminotransferase; ALT = alanine aminotransferase

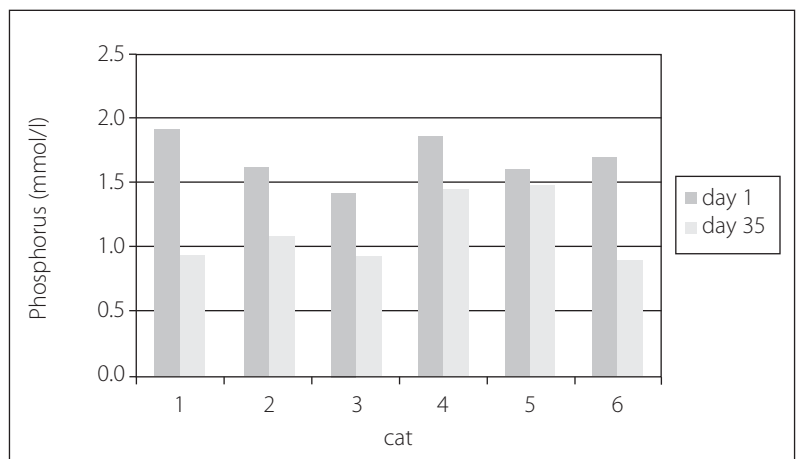
**TABLE 7:** Haematology of the 6 older cats before (day 1) and after 35 days of treatment.

period		Hb mmol/l	PCV l/l	RBC 10 <sup>12</sup> /l	MCV (fl)	MCH fmol	MCHC mmol/l	WBC 10 <sup>9</sup> /l
control	mean	7.3	0.35	8.3	42.5	0.88	20.7	21.1
	SD	1.01	0.05	1.04	1.35	0.02	0.19	6.8
treatment	mean	7.1	0.35	8.4	41.2	0.83	20.6	24.3
	SD	0.77	0.03	0.67	1.42	0.04	0.06	10.7

Hb = haemoglobin, PCV = packed cell volume, RBC = red blood cell, MCV = mean corpuscular volume, MVH = mean corpuscular haemoglobin, MCHC = mean corpuscular haemoglobin concentration, WBC = white blood cell



**FIGURE 1:** Plasma urea concentration before (day 1) and after 35 days of treatment with the chitosan/calcium supplement in the older cats (mean concentrations differed significantly between day 1 and 35,  $p < 0.05$ ).



**FIGURE 2:** Plasma phosphorus concentration before (day 1) and after 35 days of treatment with the chitosan/calcium supplement in the older cats (mean concentrations differed significantly between day 1 and 35,  $p < 0.05$ ).

are frequent with such diets and may lead to a low compliance of owners. Therefore, alternative treatments are warranted that can decrease the levels of urea and phosphorus. The study shows, that feeding a dietary supplement based on chitosan and calcium carbonate was efficient in both aspects. Plasma inorganic phosphorus levels were reduced significantly. A similar result was found in rats by Baxter et al. (2000). The results of this trial showed no influence on the urine parameters. Due to the low calcium carbonate intake by the supplement the urine pH value was not changed during the treatment.

The flexible structure of the polymer chain of chitosan could be another reason for the fixation of phosphorus in the gut. This structure enables the molecule to take suitable configurations for the complexation with metal ions and also with calcium and phosphorous. This might explain the negative digestibility of calcium in the balance trial (Bernkop-Schnürch, 2000).

The underlying mechanism that is responsible for the observed shifts in phosphorus metabolism is not clear but might be related to several factors.

In a study of Zhang and Neau (2002) the degradative activities of extracellular and cell-associated portions of rat cecal and colonic enzymes, whose activities are comparable to that in the human colon, against five chitosan qualities were characterized. The effects of the molecular weight (MW) and degree of deacetylation (DD) of chitosan on its susceptibility to degradation were investigated. In addition, the degradation function of rat bacterial enzymes was compared to that of a commercially available beta-glucosidase that contains a chitinase. The results show that rat bacterial enzymes had the ability to degrade chitosan with extracellular enzymes exhibiting a more profound effect than did cell-associated enzymes. The reaction to bacterial enzymatic degradation was dependent on both the MW and DD of the chitosan sample. Those samples with a lower MW and lower DD were more susceptible substrates.

Chitosan can act as an absorption enhancer in the intestine by increasing the residence time of drugs at mucosal sites, inhibiting proteolytic enzymes, and increasing the permeability for protein and peptide drugs across mucosal membranes. Recently, it was found that chitosan is degraded by the microflora that are present in the colon. As a result, this polymer could have promising application in colon-specific drug delivery. In consequence of the physical, chemical, and biological properties, chitosan has been used in many different formulations for drug and gene delivery in the gastrointestinal tract (Hejazi and Amiji, 2003).

The apparent protein digestibility was decreased in the treated cats. Yoshimoto et al. (1995) and Razdan and Pettersson (1996) observed a decreased apparent protein digestion in a dose-dependent manner by chitosan in rats and chickens, respectively. The authors explained this effect by a lower ammonium absorption due to high bacterial growth activities, which are able to use ammonia as a nitrogen source. The significant decrease of blood urea levels in our cats might have been caused by a reduction in protein digestibility and ammonia absorption of the treated cats.

In conclusion, the treatment had a significant effect on the phosphorus- and protein digestibility in adult healthy cats. The practical implication could be an alternative treatment option for cats refusing to ingest veterinary renal diets. Reduction of hyperphosphatemia and the resulting risk for renal hyperparathyroidism is considered as one of the most important aspects of the treatment of patients with renal insufficiency. The efficacy of chitosan and calcium supplementations as additional supplement to veterinary diets in cases with severe renal failure needs further evaluation.

Additionally, investigations on the mode of action of chitosan and the consequences for the development of secondary hyperparathyroidism and for the calcium- and protein metabolism are of clinical interest.

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# A Preliminary Clinical Evaluation of Kibow Biotics,<sup>®</sup> a Probiotic Agent, on Feline Azotemia.

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## Abstract

A clinician, curious about the manufacturer's claims, examined the results of a probiotic combination marketed as Kibow Biotics<sup>®</sup> on azotemia in cats. Results indicate a decrease in creatinine levels in six out of seven patients treated (86%) even though dosing was less than the recommended amount in most cats. This study suggests that probiotic therapy is safe and effective and indicates a place for such products in management of renal failure in cats. Further study is indicated to determine optimal dosing and potential adverse side effects, and to assess which cases are most and least responsive.

## Introduction

Feline renal failure is a significant cause of morbidity and mortality in cats in the United States.<sup>1</sup> Reducing morbidity and mortality associated with renal failure is an important goal in companion animal veterinary medicine. Regular screening of geriatric cats can assist in early diagnosis.<sup>2</sup> Ascertaining the cause of renal damage may greatly assist in formulating a therapeutic plan. Sadly, many cases present in more advanced conditions. Therapy involves reducing uremic toxins, normalizing renal blood flow and blood pressure, maintaining hydration and electrolyte balance, and supporting tissue repair when possible.

Regardless of cause, it is considered desirable to reduce levels of blood urea nitrogen (BUN) and serum creatinine in renal failure patients.<sup>4</sup> Feeding reduced levels of high biological value protein in advanced failure has been the staple treatment of chronic renal failure in cats.<sup>3</sup> Dietary therapy has been shown to increase survival of feline renal failure patients.<sup>4,5</sup> Use of other agents such as phosphorus binding substances and parathyroid hormone modulation are also utilized.<sup>5,6</sup> There are divergent opinions regarding the make up of an optimal diet for feline renal failure and more data is needed to answer these questions. Feline patients may be difficult to medicate orally over long periods of time, which creates a challenge when designing clinically useful programs for chronic use. Products selected must be well tolerated as well as effective.

A novel, biological therapeutic approach involving use of probiotic bacteria is reported to decrease BUN and serum creatinine levels in pigs and rats.<sup>7,8</sup> No feline studies are currently available. Kibow Biotics<sup>®</sup> is produced by Kibow Biotech and contains a patented mixture of probiotic bacteria consisting of *Streptococcus thermophilus*, *Lactobacillus acidophilus*, and *Bifidobacterium longum*. When these probiotic bacteria are combined with prebiotics, they form functional compositions known as *symbiotics*. *In vivo* studies show that these bacteria have an affinity for many uremic toxins. Theoretically the use of such products may support healthy bowel function, break down toxic substances, eliminate excess waste materials, and enhance the patient's immune system. In animal studies involving rats with surgically induced renal insufficiency, use of this product lead to increased survival rates of 66% and 83% in treated groups compared to 33% survival rates in placebo treated rats.<sup>7,8</sup>

The author decided to challenge the company advertising and see if positive results were obtained. This study documents those findings.

## Materials and Methods

Patients with elevated BUN and serum creatinine were selected as they came into the office. No attempt to screen patients was made. Clients were advised about the product and its potential benefits. No risks were

known. Clients voluntarily bore the expense of the product and of testing monthly for three months. The manufacturer provided the product at a discount for those undertaking the study. Values of BUN, serum creatinine, body mass, diet, and general comments were collected before, and at each subsequent visit. These were recorded on an individual work sheet and graphed for evaluation. All laboratory testing was performed by Antech laboratories in the standard way.<sup>9</sup> Body weights were determined using a single digital scale for all cats.

**Results**

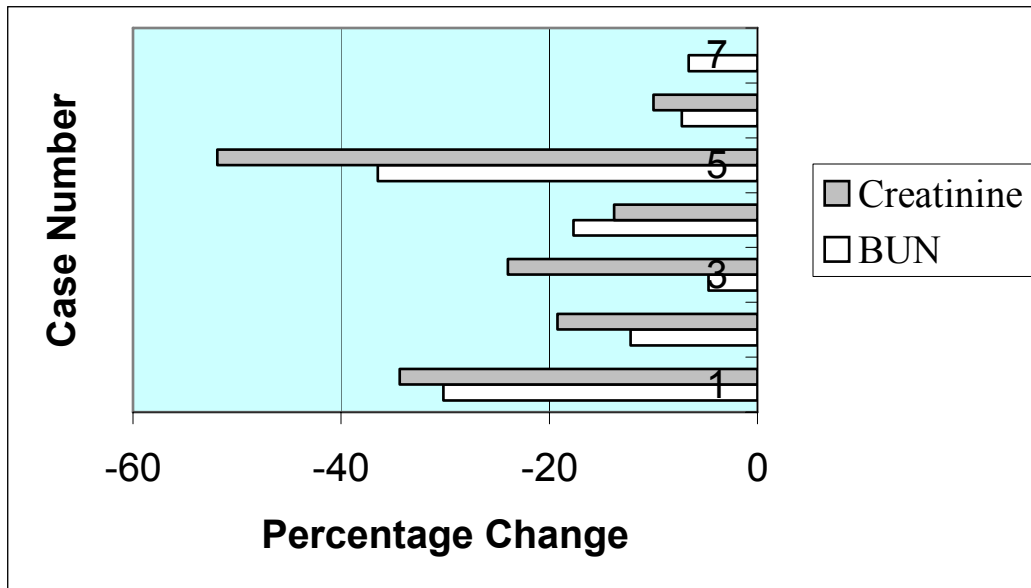
All clients presented with the manufacturer’s product data elected to be included in the study. Eight cats were enrolled over a thirty-day period. One case (number 8) dropped out after one month as the owner did not wish to have testing done monthly, was not certain that the treatment would be beneficial, and had difficulty medicating his cat. This cat was extremely uncooperative and it was agreed that dropping the treatment until more positive results were known was a good idea. All other cats completed the study. All clients have elected to continue therapy at their expense following their participation in this study. Each case response is summarized in the Table one below.

Case No.	BUN Pretx	BUN 30d	BUN 60d	% Decrease	Cr PreTx	Cr 30d	Cr 60d	% Decrease	Diet	Comments
1	43	33	30	30.2	3.2	3.1	2.1	34.4	3	Hi Protein Diet, Active
2	41	37	36	12.2	2.6	2.1	2.2	19.2	2	Difficult to dose
3	64	61	61	4.7	5.0	3.9	3.8	24.0	1	Higher vitality
4	41	ND	34	17.7	2.9	ND	2.5	13.8	2	Vomiting at whole capsule dose
5	74	51	47	36.5	7.9	4.4	3.8	51.9	1	Fibrosarcoma, Adenocarcinoma hospice; Higher vitality
6	55	45	51	7.3	3.3	3.1	3.0	10.0	1,2	Dysbiosis?, Better at ending
7	61	86*	57	6.6	2.9	3.9*	2.9	0	4	Hospitalized twice in three months <sup>+</sup>

**Table 1. Case Response Summaries.**

Legend of Abbreviations—BUN (blood urea nitrogen level reported in mg%; normal values 14-36), Cr (creatinine reported in mg%; normal values 0.6-2.4), Diet 1 (Prescription kidney diet; IVD Modified or Hill’s K/D ), Diet 2 (Commercial cat food), Diet 3 (Innova EVO), Diet 4 (home prepared meat), \* Dehydration (heat or diarrhea), <sup>+</sup> Dosing not regular.

A graphic representation of the percentage change illustrates the results below in Figure One.



**Figure 1. A Comparison of BUN and Serum Creatinine Decreases as Expressed in Percentages.**

This was a diverse group of patients. Table two summarizes other known pertinent patient factors. No attempt to alter therapy plans was made. Treatment plans were programmed based upon best therapy for that individual, and Kibow Biotic® was simply added to the cat’s program.

Case No.	Sex	Age Yrs	Wt (kg) 1	Wt (kg) 2	Wt (kg) 3	Renal Disease/ Since	Culture Urine?	Fluid Therapy	Nutritional Therapy	Homotox
1	MN	13	4.77	4.77	4.77	CGN 5 mos	No	No	No	Yes
2	MN	15	6.68	6.51	6.16	CGN	Neg	No	Yes, 2	No
3	FN	18	3.13	2.93	2.93	CGN 3 yrs	Neg	No	Yes, 1,2,3	No
4	MN	13	5.68	ND	5.65	FUS related Post-renal Blockage 1.5 yrs	No	No	Yes 1	No
5	FN	17	8.44	8.81	9.31	CGN Paraneoplastic Tubular dz? 1.5 yrs	Neg	Yes SQ	Yes 1, 2	Yes
6	MN	10	4.95	4.83	4.94	CGN 4 mos	Neg	Yes SQ	Yes 2	No
7	FN	19	2.59	2.81	3.1	Recurrent Pyelonephritis 3 yrs	No	Yes IV I/P SQ O/P	Yes 1, 2	Yes

**Table 2. Patient Factors.**

Legend of Abbreviations—Kg (kilogram), M (male), N (neutered), F (female), CGN (chronic generalized nephropathy), dz (disease), FUS (feline urologic syndrome), IV I/P (intravenous fluids in-patient), SQ O/P (subcutaneous fluids out-patient), Nutritional therapy 1 (diet), Nutritional therapy 2 (Standard Process Feline Renal Support), Nutritional therapy 3 (Renagen®, a Chinese herbal by Thorne Labs), Homotox (Homotoxicology, a specialized form of homeopathic therapy).

Cats received either one-half capsule twice daily or one capsule daily. Clients admitted to varying this dosage so a valid comparison based upon dose or route could not be made.

<b>Weight, Kg</b>	<b>Morning Dose</b>	<b>Evening Dose</b>
< 1 kg	1	0
1-2 kg	1	1
2-4 kg	2	1
4-8 kg	2	1-2

**Table 4. Kibow Biotech’s Company Recommendations for Dosing of Veterinary Patients.**

Most patients in this study took one capsule daily or half capsule twice daily mixed with food. This is in disagreement with the company’s advise to give the capsule whole or with liquid food.<sup>10</sup>

## Discussion

Examination of the limited data above shows a very clear relationship between use of the probiotic and decreasing azotemia. 7/7 (100%) of cases showed a decrease in BUN, which varied from 4.7% to 36.5%. Creatinine also showed improvement with 6/7 (85.7%) showing decreases ranging from 10% to 51.9%. Such a decline must originate from altered blood flow, reduced toxin presentation, increased toxin excretion/conversion, or other unknown issues. Kibow Biotech coined and trademarked the term, Enteric Dialysis™ to describe the removal of uremic toxins by bacterial action in the colon.<sup>10, 11</sup> This study supports that theory, even though the dosing was lower than that recommended by the company.

It would have been interesting to monitor other parameters such as packed cell volume, and phosphorus, but finances limited the scope of this particular study. Body weights generally fell in 4/7 (57.1%) over the 60-day period of this study. The exceptions to this were three cases that either maintained weight, 1/7 (14.3%), or that gained weight, 2/7 (28.6%). These three cats all received homotoxicology support for their kidneys in addition to their other treatments, which raises an interesting opportunity for further study in attempting to determine the best protocols for renal failure in cats. Reports of homotoxicology benefiting renal disease in veterinary patients exist.<sup>12,13,14,15</sup>

Case number seven deserves comment. This is a very aged cat that was hospitalized three times in the last year for chronic renal failure, dehydration and recurring bacterial pyelonephritis. She has very advanced oral cavity disease, the owner is reluctant to do dental prophylaxis and she had received antibiotics for long periods of time. Chronic antibiotic therapy can damage intestinal flora and may affect negatively the immune system of the patient.<sup>16, 17</sup> During a three-month period of time her serum creatinine varied from 3.9 mg/dl to 2.6 mg/dl and back up again. Her pre-hospitalization serum creatinine was 3.9 mg/dl and she completed her study period with a serum creatinine of 2.9 mg/dl, which represents a substantial improvement in a case this severe. Her serum creatinine and BUN did show declines but the study overlapped one of her decompensations and hospitalizations, so it appears that little actually happened. However, the cat has shown amazing increases in appetite and energy per the owner’s report as well as experiencing weight gain when it was not expected. This cat did experience some diarrhea that resolved and may be related to intestinal dysbiosis and/or gastrointestinal cleansing resulting from improved intestinal floral health. During periods of bacterial die-offs toxins may be released as the body flushes these toxins out the gastrointestinal system. The diarrhea seemed to be associated with antibiotic therapy and worsening of the azotemia. The exact reasons for this cannot be known for certain.

Case number five has been diagnosed with vaccine-related-fibrosarcoma and adenocarcinoma for a year-and- a-half. This client has elected modified hospice for this patient, and per her caregiver, her quality of life has improved markedly since taking the probiotic. She is very active and feels well even though her tumor is relatively large. This case has not had kidney biopsies but it is likely that she has some issues associated with the advanced state of her tumor involving glomerular/tubular function.

All cats in this study have palpably small kidneys indicating chronic disease. Case four suffered a rather bad case of FUS blockage several years ago and it is suspected that this contributed to his present renal failure. All cats in this study received vaccinations. Our clinic utilizes a reduced vaccination protocol. We vaccinate for FVRCP only every three years, and we cease vaccinations in indoor cats with other disease states in support of vaccine label recommendations.

Patient tolerance of the product was good, but it was difficult to give the recommended amounts without mixing it with food. No cats would accept it as a capsule and several clients balked at the cost. We attempted to put it in water and drench these patients but this was not accepted. Finally, all cats ingested it easily from canned cat food. There is some concern that giving probiotics with food may decrease their numbers, but judging from the above results there is a clinically significant effect from ingesting adequate numbers of these bacteria. Higher dosing may give even better results. Only one cat suffered any discomfort, and that was case number four. This cat experienced vomiting soon after receiving the oral product mixed with water. The owner ceased the product for three days and began mixing it with food and the cat did fine after this.

Veterinarians practicing Complementary and Alternative Veterinary Medicine (CAVM) have long proposed and advocated the use of probiotics in aging veterinary patients. The alternative movement has frequently been criticized for use of such nonvalidated procedures but they frequently turn out to be very useful. CAVM has reported a wide number of benefits observed clinically. This study demonstrates another value to this practice and validates those clinical opinions. Hopefully, other practitioners will do similar small clinical reports that can be used to target more precisely those modalities that are particularly promising for treatment of feline azotemia and renal failure. Immediate opportunities exist in comparing various probiotic products for activity in reducing azotemia. Kibow Biotech states that the specific strains of bacteria used in Kibow Biotic<sup>®</sup> are more effective than other products currently on the market. It would be very useful to compare various other CAVM practices as part of a protocol to treat chronic renal failure in cats.

The author has personally seen many remarkable improvements in cats suffering from renal disease after administering nutraceuticals, whole food and glandular supplementation, Traditional Oriental Medicine, homotoxicology, biopuncture, and homeopathy. Other approaches exist using western herbs as well. Integrative Veterinary Medicine is gaining momentum and this would seem a very fruitful area for research and validation of modalities potentially helpful to feline medicine. Of even greater interest is the possibility of reducing renal failure, prolonging the period of normalcy, and even potential life extension by early intervention with probiotic formulas as medicants or food additives. The biological therapy movement would receive such research data with great excitement.

In conclusion, Kibow Biotics<sup>®</sup> seems to have benefited these cases. The manufacturer's promise of decreasing azotemia appears to be verified, and these patients did experience improved health and vitality. This data does not refute or substantiate improved immune status or longevity. The cats generally did have good stools and only one case of vomiting occurred. Clients were happy to use the product and have been pleased to reorder indicating client satisfaction and ease of use. The single case of diarrhea resolved and did not recur. The author would like to see more involved placebo-controlled, double-blind trials comparing this product to other probiotic formulations.

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