

Dried Solidified Blood Calculi in the Urinary Tract of Cats

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We have noted an increased number of calculi submitted to the Gerald V. Ling Urinary Stone Analysis Laboratory, University of California, Davis, that do not contain crystalline material but appear to be composed of dried solidified blood (DSB). Canine and feline laboratory records from 1986–2003 were reviewed for samples composed of >99% DSB. No calculi from dogs were found, but specimens from 49 cats were composed of >99% DSB, of which almost half ($n = 22$) had been submitted after 2001. The DSB calculi had been removed surgically or by postmortem examination from all areas of the upper and lower urinary tract. All samples were well formed and could be divided in half with Rongeur forceps. Detailed case information was available for 12 cats. Urinalyses were available for 9 of the 12 cats, and the mean specific gravity was 1.017 (range, 1.009–1.032). Red blood cells were reported in the sediment of all cats, with most containing >100 RBC/hpf. Ureteral obstructions but no radio-opaque calculi were visible by radiography, including radiographic contrast studies. Reports of ultrasound examinations were available for 10 cats, and discrete calculi were not recorded. In addition to polarized light microscopy, infrared spectroscopy and electron probe microanalysis were performed on a subset ($n = 6$, DSB calculi; $n = 3$, control calculi) of samples. Significantly more carbon, nitrogen, and sulfur ($P = .012$, $P = .02$, and $P = .012$, respectively) were present in the elemental analysis of the DSB calculi than in the control calculi, suggesting that the DSB calculi are primarily formed from organic material. At this time, we are uncertain why these DSB calculi become solidified, and we recommend that samples be submitted both in formalin and preservative free to further investigate their etiology.

Key words: Cat; Bladder; Lower urinary tract disease; Ureters.

Signs of lower urinary tract disease in cats, including stranguria, dysuria, pollakiuria, and urinating in inappropriate places, can be caused by a variety of disorders, including urinary tract infections, cystic calculi, neoplasia, and idiopathic cystitis. In approximately 15–21%^{1,2} of cats presenting with clinical signs related to the lower urinary tract, the clinical signs are attributed to urinary calculi. By definition, a calculus is an abnormal concretion occurring within the animal's body and is usually composed of mineral salts.³ Urolithiasis is a well-recognized problem in cats and can account for up to 21% of cats with lower urinary tract signs. The two most common stone types contain calcium oxalate (CaOx) and struvite. Several other stones types, such as urate and cystine, are also reported in cats.^{4,5} The bladder is the most common site for urinary calculi in cats; however, recent reports note an increased number of uroliths obtained from the upper urinary tract.^{6,7} In addition to the more common calculi mentioned above, during the past several years, we have noticed an increasing number of samples submitted to our laboratory as urinary calculi that do not contain crystalline material but appear to be composed of dried solidified blood (DSB). These samples are not gelatinous blood clots. The purpose of this study and report was to better characterize these DSB calculi and to present

relevant clinical data and the morphologic characteristics of these samples.

Materials and Methods

Records from the Gerald V. Ling Urinary Stone Analysis Laboratory, School of Veterinary Medicine, University of California, Davis, between 1986 and 2003 were reviewed to find samples from cats and dogs that contained noncrystalline material as the primary component. Only samples composed of <1% crystalline material or samples in which the core was composed of <1% crystalline material were included in the study. Samples that had small amounts of dried blood on the surface of the calculus were not evaluated. However, 3 specimens of DSB calculi that also contained calcium phosphate (CaP) or CaOx in the surface, inner layer, or core served as controls. The purpose of these controls was to make certain that subsequent infrared (IR) spectroscopy and electron probe microanalysis (microprobe) would detect the mineral components when those crystalline components were present in the unknown specimens.

Specimens submitted to the Stone Analysis Laboratory had been accompanied by information from the referring veterinarian that included the patient's age, sex, and breed, as well as the location from which the sample had been obtained, whether the urine had been cultured and results of that culture, and whether the patient had a history of previous stone formation. Records from patients seen at the University of California, Davis, Veterinary Medical Teaching Hospital (VMTH) were reviewed for additional clinical and diagnostic information.

Qualitative and quantitative analyses of the calculus specimens we obtained included the use of polarized light microscopy, IR spectroscopy, microprobe, and histology. First, all samples received by the laboratory were weighed and measured, and the external physical features (eg, color, shape, and surface texture) were determined with a stereoscopic dissecting microscope.⁸ Each sample was cracked with Rongeur forceps; and the internal features were examined with a dissecting microscope, noting the presence of any visible layering.

Polarized Light Microscopy. Samples from each visible layer were examined separately by the oil immersion method of optical crystallography with a polarized light microscope^b to determine the mineral composition and to estimate the relative volumetric percentages of each layer.^{8,9}

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Submitted August 4, 2005; Revised October 11, 2005; Accepted December 12, 2005.

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0891-6640/06/2004-0005/\$3.00/0

IR Spectroscopy. A subset of 9 specimens (6 DSB and 3 control samples) were analyzed with a Fourier-transform infrared spectrometer^c equipped with OMNIC spectroscopy software and the Kidney Stone Library and Analysis software.^d The 6 DSB specimens were chosen randomly from those samples that had enough material to perform the subsequent analyses. The 3 control samples were also chosen randomly from the samples that contained CaP or CaOx, in addition to the DSB revealed by polarized light microscopy. Pellets of each layer of each specimen were prepared at a concentration of 1 part finely ground specimen to 100 parts of potassium bromide (KBr)^e and were then pressed with a KBr die and a carver hydraulic press^f at 44,000 psi under vacuum for 2 minutes. The pressure was then released, and the pellet was pressed again for 1 minute. The pellet was then placed in a magnetic KBr pellet holder^g and scanned with a KBr beam splitter and a DTGS KBr detector.^h The OMNIC Kidney Stone Library and Analysis software^h was then used to match the values from the search results and calculate the concentrations of the components in the unknown samples.

Microprobe. A subset of samples ($n = 9$; the same samples analyzed by IR spectroscopy) were analyzed qualitatively and quantitatively with an electron microprobe analyzerⁱ equipped with 1 energy-dispersive spectrometer and 5 wavelength-dispersive spectrometers. For qualitative analysis, the samples were placed on double-stick carbon tape and coated with approximately 250 angstroms of carbon to make them conductive. Interpretation of the results from the energy-dispersive spectrometer revealed all possible elements that may be present in the sample over an area of approximately 20 μm across and 10 μm deep on the rough surface of the sample. The elements for quantitative analysis were selected on the basis of the results from the qualitative studies. For quantitative analysis, the calculus samples were mounted in epoxy, cut, and polished to provide a smooth surface; the fresh surfaces were also coated with approximately 250 angstroms of carbon to give them a conductive surface and were then analyzed as previously described.¹⁰ The technician performing the qualitative and quantitative analyses was unaware of the results of the previous polarized light microscopy and IR analyses.

Histopathology. Three calculi were submitted for histological analysis. They were placed in formalin and routinely processed and stained with hematoxylin and eosin.

Statistics. The quantitative elemental analysis results from calculi containing DSB and from control calculi containing crystalline material were compared by an exact Mann-Whitney test. Results with a P value $<.05$ were considered statistically significant.

Results

Clinical Case Results

From 1986–2003, the Stone Analysis Laboratory received 4,933 samples from cats and 21,784 samples from dogs for evaluation. Forty-nine cats had specimens composed of $<1\%$ crystalline material. Almost half ($n = 22$) of these calculi were submitted after 2001. Most cats were domestic short- or long-haired, except for 2 Maine Coons, 2 Siamese, 1 Himalayan, 1 Snowshoe, 1 Ragdoll, and 1 Balinese. The mean age of the cats was 9 years (range, 1–15 years). Thirty-three of the cats were castrated males, and 13 were spayed females. Two male cats and 1 female cat were intact.

No calculi containing $<1\%$ crystalline material were found in samples from dogs. Only 3 calculi containing any DSB were identified from dogs. Two of these calculi contained particles of dried blood rather than the actual

well-formed solid concretions we encountered in the cat samples, and 1 contained only a small amount of DSB in the core while the rest of the stone was composed of CaOx. Therefore, these calculi did not meet the inclusion criteria for our review.

The calculi had been removed surgically from all cats, except for 3 where the samples had been obtained during the postmortem examination. On the basis of the submittal information, the DSB calculi had been removed from a variety of locations in the urinary tract, including the bladder ($n = 20$) or bladder and urethra ($n = 3$), urethra ($n = 5$), left renal pelvis ($n = 4$), right renal pelvis ($n = 5$), right ureter ($n = 4$), left ureter ($n = 3$), right renal pelvis and ureter ($n = 1$), left renal pelvis and ureter ($n = 1$), both renal pelvises ($n = 1$), and voided ($n = 1$); 1 sample submitted to the laboratory did not have a location listed on the questionnaire. Results of urine cultures were reported for 35 cats: 3 specimens grew *Escherichia coli*, 2 grew *Bacillus spp.*, and 1 grew *Staphylococcus spp.* Only 2 cats had their calculi cultured, and both resulted in no growth. Only 2 cats had historical urolithiasis reported on the questionnaire, and these analyses were not available for review.

For 12 cats that were presented to the VMTH, additional clinical history was available. Only 4 calculi had been located in the lower urinary tract, whereas all others had been surgically removed or obtained at the postmortem examination from the ureters, renal pelvis, or both. Seven of the ureteral calculi were submitted in the year 2000 or later. Six of the 12 cats had presented with acute or “acute on chronic” renal failure because the calculi were causing ureteral obstruction. Urinalyses were available for 9 of 12 cats, and the mean specific gravity was 1.017 (range, 1.009–1.032). Red blood cells were reported in the sediment of all cats, usually >100 RBC/hpf. One cat had concurrent diabetes mellitus.

Plain radiographs were available for review in 7 cats. Three cats had radio-opacities noted in the kidneys that were thought to be renal mineralization, although no defined stones were reported. Renomegaly was noted unilaterally in 3 cats and bilaterally in 1 cat. Ultrasound reports were available for review in 10 of the 12 cats. Eight of the reports noted renal pelvic dilation ranging from mild to severe. Three reports mentioned a small contralateral kidney. Three ultrasound reports also noted ureteral dilation, and 1 noted free fluid within the abdomen. Ureteral obstruction was suspected in all cats, but no discrete calculi were noted. A blood clot was reported in the bladder of 1 cat, in addition to a ureteral obstruction. Contrast radiography was reported in 5 of the 12 cats. Interpretation of one study revealed a right proximal ureterolith by nephropyelogram, and the others documented ureteral obstructions but no defined calculi. One positive contrast cystogram reported non-radio-opaque calculi within the bladder lumen. Only one cat had computed tomography performed; interpretation revealed moderate left renomegaly and hydroureter. A ureteral obstruction was suspected, but no defined stone could be found. In that same cat, the right kidney was small, and a stone was noted in the right renal pelvis.

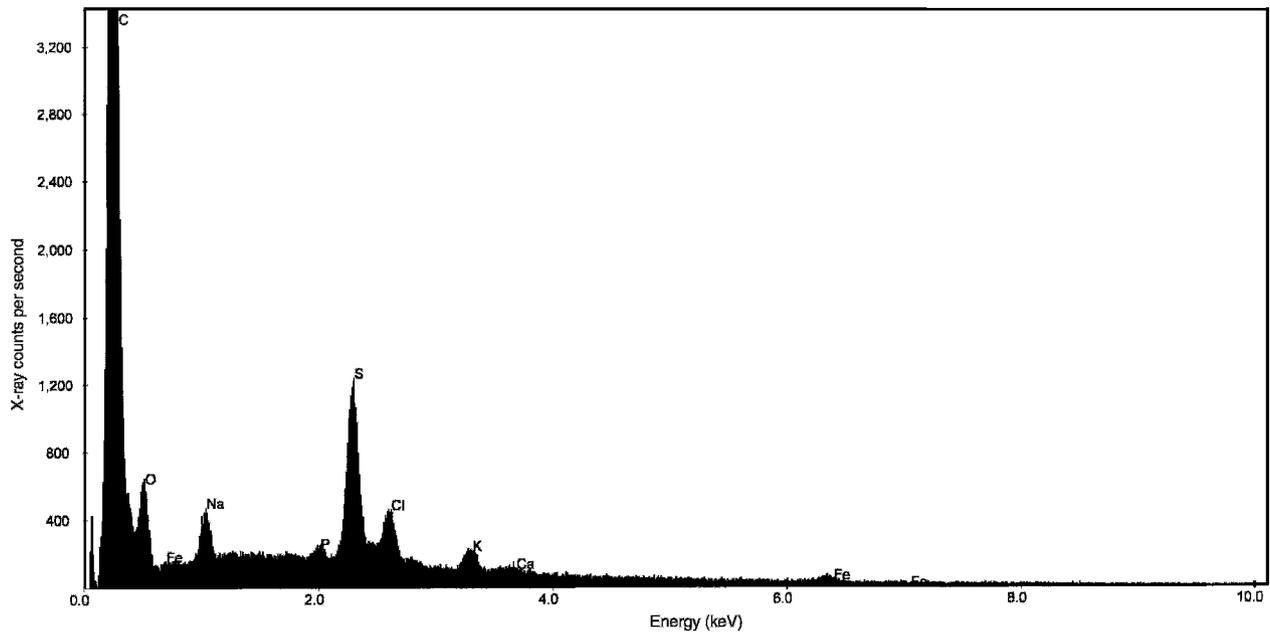


Fig 1. Representative qualitative microprobe analysis from a dried solidified blood calculus. Interpretation of the results from the energy-dispersive spectrometer reveal all possible elements that may be present in the sample over an area of approximately 20 μm across and 10 μm deep on the rough surface of the sample. Carbon (C), oxygen (O), iron (Fe), sodium (Na), phosphorus (P), sulfur (S), chlorine (Cl), potassium (K), and calcium (Ca). The high qualitative C peak is partially due to the carbon coating.

Gross Specimen Analysis

The samples were round or irregularly shaped and were firm to very hard. Forty-five of the 49 samples had measurements and weights recorded. The average weight of the samples was 0.065 g (range, 0.01–0.51 g). The average size was 49.2 mm^3 (range, 0.48–386.8 mm^3). Both larger and smaller samples were identified in all locations in the urinary tract. The average number of samples submitted to our laboratory per cat was 5 (range, 1–17). Thirty-six of the cats had only DSB calculi submitted to the laboratory. The remaining cats had primarily DSB calculi but also some CaOx ($n = 8$), CaP ($n = 3$), CaOx and CaP ($n = 1$), or struvite ($n = 1$) calculi submitted. Thirty-nine of the calculi appeared to contain only 100% DSB. In the DSB calculi that contained other mineral types, 5 contained 1% CaOx and 3 others contained 1% CaP. One sample contained a core of 100% DSB and an outer shell of 50% CaOx and 50% CaP, and 1 DSB calculi had a small amount of struvite crystals present within the sample.

Results of Polarized Light Microscopy

All samples appeared to be composed of DSB on gross examination but were not blood clots. All samples were well formed and could be divided in half with Rongeur forceps. When the internal morphology of the samples was examined with a dissecting microscope, differences in porosity were observed. Five were extremely dense, and the cut surface resembled polished obsidian; whereas others ($n = 25$) exhibited more porosity and an unoriented appearance throughout the interior of the calculi. The remaining samples were too small to document the internal morphology. When each

layer of the sample was examined with polarized light microscopy, no mineral crystals were observed, except in the 3 control samples. Under direct light, microscopic examination of the calculus material revealed DSB without the presence of crystalline material. In the specimens which contained DSB in addition to minerals, the mineral crystals were readily detected and identified with polarized light microscopy and reference standards,⁹ and their identities were confirmed by additional methodologies.

IR Spectroscopy

When IR spectroscopy was performed on a subset of 6 specimens with no crystalline material identified when examined by polarized light microscopy, the computerized kidney stone library software indicated that no match for the calculus material could be found in the kidney stone library computer database. When IR spectroscopy was performed on the 3 control samples containing either CaP or CaOx (identified by polarized light microscopy) in addition to the DSB, the CaP or CaOx were readily identified by the kidney stone library software.

Microprobe

Qualitative and quantitative microprobe analyses were then performed on the same subset of 6 specimens with no crystalline material identified when examined by polarized light microscopy and IR spectroscopy. The following qualitative elemental peaks were present in all 6 specimens: carbon (C), oxygen (O), sodium (Na), sulfur (S), chlorine (Cl), potassium (K), and calcium (Ca) (Fig 1). In addition, magnesium (Mg) peaks were present in 4 of the 6 specimens, and phosphorus (P)

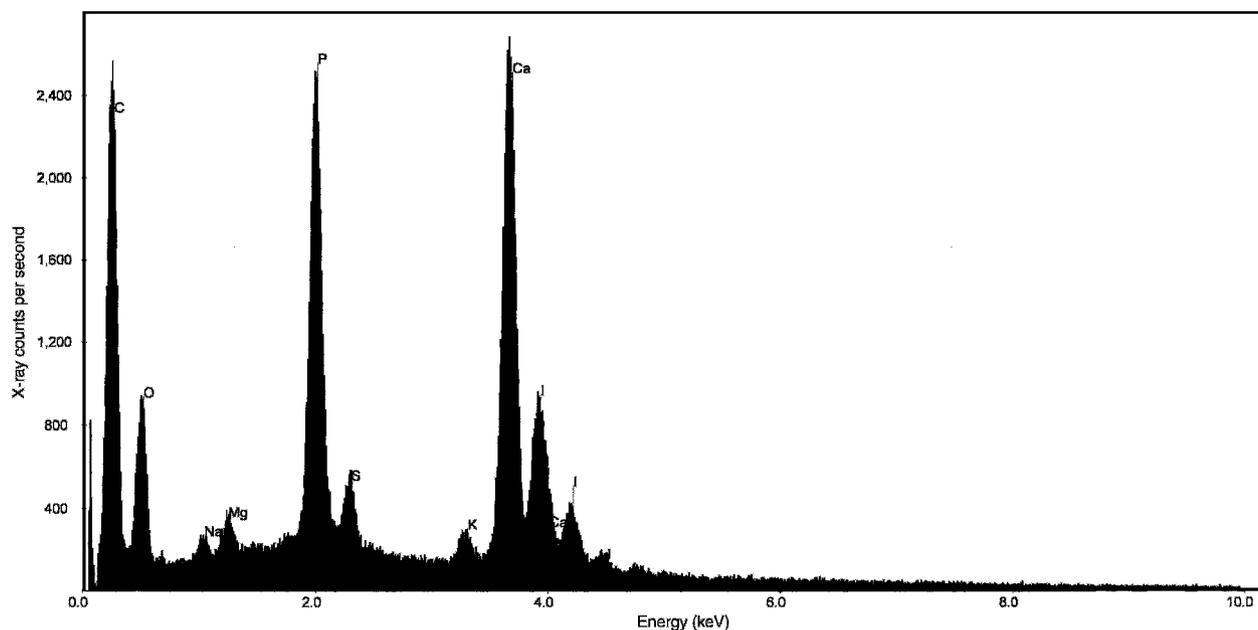


Fig 2. Representative qualitative microprobe analysis from a control sample. Interpretation of the results from the energy-dispersive spectrometer reveal all possible elements that may be present in the sample over an area of approximately 20 μm across and 10 μm deep on the rough surface of the sample. Carbon (C), oxygen (O), sodium (Na), magnesium (Mg), phosphorus (P), sulfur (S), potassium (K), calcium (Ca) and iodine (I). Note the larger peaks for Ca and P. This control sample contained calcium phosphate, which was detected by polarized light microscopy and infrared spectroscopy.

peaks were present in 3 of the 6 specimens. One sample contained a silica (Si) peak, and 1 also contained a peak for iron (Fe).

The control specimens had peaks for either Ca only or Ca and P that were much higher than in the other 6 specimens (Fig 2). The following elements were also present in the samples: C, O, Fe, Na, Mg, Si, P, S, Cl, and K. One control sample also had an aluminum peak not found in any of the other specimens. Another control sample also contained a primary and secondary iodine (I) peak.

In the quantitative microprobe analysis (quantified by weight percent), all of the elements detected in the qualitative analyses of the 6 DSB calculi and the 3 control calculi were observed to be present. Two of the control calculi had distinct areas noted in their cross sections, and each area was analyzed separately by the technician (dark and light areas containing noncrystalline and crystalline material, respectively). In the 5 specimens containing no crystalline material, all the elements (Na, F, Mg, Al, Si, P, S, Cl, K, Ca, and Fe) were present in trace ($\leq 1\%$) amounts. There was significantly more C, N, and S in the samples where polarized light microscopy and IR spectroscopy did not detect any crystals ($P = .01$, $P = .02$, and $P = .01$, respectively; Table 1). Significantly more Ca, P, and O were found in the samples where crystalline material was previously noted ($P < .01$, $P = .04$, and $P = .01$, respectively). No significant differences were noted in any of the other elements analyzed.

In one DSB calculus, the following elements were detected in the qualitative analysis: C, O, Na, Mg, Si, P,

S, Cl, K, Fe, and Ca. The quantitative analysis of this sample had higher concentrations of Ca and P than the other 5 samples, but the concentrations were not in the ranges observed in the crystalline control samples. The concentrations of the remaining elements in this sample were noted in trace amounts, with the exception of C, O, and nitrogen (N).

In the control specimens containing CaP or CaOx, distinct phases within the interior of the calculi were observed in the quantitative analyses. The phase containing the CaP or CaOx had a much higher concentration of either Ca and P or just Ca, respectively, than the second phase where no crystalline material was previously identified. The noncrystalline phase (presumed to be DSB) had trace ($\leq 1\%$) to minor concentrations (1–5%) of all of the elements present, except C and O and in some cases N.

Histopathology

Histological features of the 3 calculi were consistent with a coagulum of red blood cells in which individual cells generally lacked distinct borders, although occasionally individual erythrocyte morphology was appreciated. In a sample where crystalline material was previously identified, refractile crystalline material was scattered multifocally throughout the coagulum, and small rafts of degenerate epithelial cells were inconsistently adherent the surface of the coagulum.

Discussion

Although by definition calculi usually contain crystals,³ we have called these samples DSB calculi even

Table 1. Quantitative electron probe microanalysis. The elements for quantitative analysis were selected on the basis of the results from the qualitative studies. In 5 specimens containing no crystalline material, all of the elements (Na, F, Mg, Al, Si, P, S, Cl, K, Ca, and Fe) were present in trace amounts ($\leq 1\%$) amounts. There was significantly more C, N, and S in the samples where polarized light microscopy and IR spectroscopy did not detect any crystals ($P = .01$, $P = .02$, and $P = .01$, respectively). Significantly more Ca, P, and O were found in the samples where crystalline material was previously noted ($P < .01$, $P = .04$, and $P = .01$, respectively). No significant differences were noted in any of the other elements that were analyzed. DSB (dried solidified blood).

Element	Elements (weight percent) in DSB calculi (Mean \pm SD) N=6	Elements (weight percent) in Control calculi (Mean \pm SD) N=3	P value
Al	0.44 \pm 0.52	0.11 \pm 0.14	0.26
Cl	0.21 \pm 0.12	0.07 \pm 0.06	0.08
F	0.20 \pm 0.53	1.25 \pm 1.83	0.04
Fe	0.09 \pm 0.08	0.05 \pm 0.04	0.68
K	0.09 \pm 0.07	0.21 \pm 0.22	0.52
Mg	0.27 \pm 0.19	0.53 \pm 0.70	0.78
N	6.56 \pm 3.76	0.46 \pm 0.70	0.02
Na	0.44 \pm 0.47	0.38 \pm 0.36	0.81
P	1.04 \pm 2.77	6.42 \pm 9.43	0.03
Si	0.01 \pm 0.01	0.00 \pm 0.00	0.15
C	0.21 \pm 4.58	0.07 \pm 1.84	0.01
Ca	2.30 \pm 4.90	30.9 \pm 1.3	<0.01
O	21.2 \pm 4.2	45.23 \pm 6.89	0.01
S	1.35 \pm 0.33	0.16 \pm 0.06	0.01

though no crystalline material was found. During the same time period that we analyzed these samples from cats, we did not identify any DSB calculi from dogs meeting the inclusion criteria. Therefore, these DSB calculi analyzed at our laboratory appear to be primarily a problem in cats. Moreover, they appear to be increasing in incidence because more than half of the samples were submitted during the past 5 years. The number of submittals of feline calculi to the Stone Analysis Laboratory has not changed significantly during the past 15 years.

Very little information pertaining to DSB calculi was found in the literature. Osborne et al^{11,12} describe "blood clots mineralized with calcium phosphate." However, many of the calculi we analyzed did not contain any mineral component, and when present, crystalline material consisted not only of CaP but also CaOx and rarely struvite.

It also has been suggested that DSB calculi come primarily from the renal pelvis and less commonly from the lower urinary tract.⁵ Although the calculi we analyzed from cats submitted by the VMTH were primarily removed from the upper urinary tract, 57% of the calculi from cats submitted by referring veterinarians were removed from the bladder, urethra, or both. Therefore, these calculi can be found anywhere in the urinary tract. The difference in case distribution between the VMTH and private practitioners likely reflects the referral of cats to the VMTH that require complex surgeries or specialty medical care.

A variety of imaging studies were performed on the VMTH cases, and these calculi did not appear radiodense in most cases. Moreover, no discrete calculi were identified on ultrasound examination. Contrast studies did suggest obstruction (renomegaly and hydronephro-

sis) but rarely identified calculi. This suggests that when an upper urinary tract obstruction is present but no radiodense calculi are noted, DSB calculi should be considered as a cause.

From the analyses we performed, it is not possible to determine how the DSB calculi formed in the cats. Similar to results in previous reports,¹² no evidence of bilirubin was detected even though many of these calculi appeared black. Several of the animals had historical hematuria, and it is possible that macroscopic or microscopic renal or bladder hematuria contributed to DSB calculi formation. Because of the retrospective nature of this paper, it is possible that other cats had hematuria that was not reported on the submittal form or in the patient record. However, hematuria alone would not entirely explain the formation of these DSB calculi, and not all cats with hematuria form DSB calculi. A cause-and-effect relationship cannot be determined from this study. Although only 3 urine cultures were positive for bacteria, only 2 calculi were cultured; and a bacterial urinary tract infection could have contributed to the formation of these calculi. Osborne et al¹² suggest that formation of highly concentrated urine with gross hematuria might contribute to the formation of mineralized blood clots with CaP. The mean urine specific gravity for the cats we studied was only 1.017; however, we were unable to obtain urine samples at the time the DSB calculi were actually formed.

These samples were very firm and stone-like and were not characteristic of blood clots. These calculi composed entirely of DSB, reported here, are solid well-formed concretions that can be cracked in half with Rongeur forceps—this is an important distinction between the DSB calculi and the blood clots that might be removed

during surgery or the calculi with large amounts of blood coating the surfaces. All of our diagnostics failed to identify any crystalline material in the majority of samples. It is possible that the DSB calculi could have dried out and hardened after removal from the cat. However, one of the authors (AEK) confirmed that the samples were very firm immediately after removal from the urinary tract. Furthermore, all samples were submitted to the Stone Analysis Laboratory, and we presume that referring veterinarians also thought the samples were crystalline in nature.

For most specimens submitted, optical crystallography is sufficient to identify the crystalline mineral components of the calculi. Because the refractive indices for the common mineral components of urinary calculi have been established, the refractive indices of the crystals in the calculus specimen can be compared with the established reference indices, and a match for the crystals can be determined. No crystals could be identified with polarized light microscopy in most of the samples we presented in this study. Because we saw no crystalline material in these DSB calculi, IR spectroscopy was performed on a subset of 6 specimens to evaluate these calculi for minerals not typically noted. No match in the computerized kidney stone library software was found, providing more support for the conclusion that no crystalline material was present within these samples.

Because no crystalline material was identified by IR spectroscopy, these selected samples were analyzed by qualitative and quantitative microprobe analysis. In addition, we performed qualitative and quantitative microprobe analysis on a subset of 3 control specimens to verify the presence of either CaP or CaOx. If the 6 specimens of calculi presumptively identified as containing only DSB had also contained these minerals, then the elemental peaks should have been much higher for Ca and P or just Ca than the elemental peaks that were present in the qualitative microprobe analyses of the calculi composed entirely of DSB. If struvite had been present in the specimens of DSB calculi, then the quantitative analyses would have indicated major amounts of Mg (as well as P); however, when Mg was present in the DSB samples, it was only present in trace to minor amounts.

Quantitative analyses of the DSB calculi indicated there was significantly more C, N, and S and trace-to-minor concentrations of all of the additional elements present throughout the cross sections of calculi examined, which is consistent with the calculi being composed primarily of organic material rather than crystalline minerals. Although the samples are coated with C, the total weight percent of C present in the quantitative analysis of the DSB specimens represents the actual C content of the specimen because the standardized amount of C in the coating was subtracted.

Feline red blood cells are also very high in sulfur content, which could explain the higher concentrations of sulfur we noted. We do not believe that the presence of S in these calculi is because of a sulfonamide component. Sulfonamide crystals that can precipitate

and form components of urinary calculi can be readily observed by optical crystallography, and no sulfonamide crystals were observed in any of the DSB calculi or DSB layers of the control calculi. One sample contained Al in the qualitative analysis. It is unclear at this time why this element was present, but no history was available to identify any Al-containing medications in this cat's medical record. The high weight percentage of oxygen noted in the control samples is likely because of the high oxygen content of calcium oxalate monohydrate and apatite ($\text{CaC}_2\text{O}_4 \cdot \text{H}_2\text{O}$ and $\text{Ca}_5(\text{PO}_4)_3$, respectively).

No samples were submitted in formalin, so it was difficult to perform histological analyses, which is why it was attempted in only a few samples. Because the calculi were not fixed with the formalin preservative, we were unable to test them for other possible constituents in the matrix by immunohistochemical analyses. Some epithelial cells were noted in the samples, and just like other calculi, some renal, ureteral, or urothelial cells can become adhered or even trapped in the matrix. If surgeons remove multiple calculi that appear similar to the DSB calculi we have described, we recommend placing some in formalin and submitting others for routine stone analysis. If all samples are placed in formalin, then crystalline material may not be properly identified.

These findings substantiate the fact that the DSB samples we analyzed contained very little, if any, mineral components commonly found in urinary calculi and appeared to be composed primarily of organic material. Although we are still uncertain how these samples form *in vivo*, further analyses and complete histories will be beneficial to formulate proper prevention strategies. Currently, it seems prudent to recommend additional fluid intake and search for causes of inflammation and ischemia that could contribute to hematuria.

Footnotes

- ^a Model 569, American Optical Co, Buffalo, NY
 - ^b Universal polarizing microscope, Carl Zeiss Inc, Thornwood, NY
 - ^c Impact 410 Spectrometer, Nicolet Instrument Corporation, Madison, WI
 - ^d Nicolet Instrument Corporation, Madison, WI
 - ^e Thermo Electron Corporation, Asheville, NC
 - ^f Carver Hydraulic Unit Model #3912, Wabash, IN
 - ^g Specta-Tech Inc, Shelton, CT
 - ^h Nicolet Instrument Corporation, Madison, WI
 - ⁱ SX-100 electron microprobe analyzer, CAMECA, Courbevoise, France
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Acknowledgments

The authors wish to thank Dr Phil Kass (Department of Population Health and Reproduction, University of California, Davis, CA) for his assistance with statistical analyses, and Sarah Roeske (Department of Geology, University of California, Davis, CA) for her expertise in microprobe analysis.

References

1. Kruger JM, Osborne CA, Goyal SM, et al. Clinical evaluation of cats with lower urinary tract disease. *J Am Vet Med Assoc* 1991;199:211–216.
2. Buffington CA, Chew DJ, Kendall MS, et al. Clinical evaluation of cats with nonobstructive urinary tract diseases. *J Am Vet Med Assoc* 1997;210:46–50.
3. Taylor EJ. *Dorland's Illustrated Medical Dictionary*. Philadelphia, PA: WB Saunders; 1988.
4. Westropp JL. Epidemiology of feline urolithiasis. In: *Feline Urinary Calculi: The New Stone Age*. Montepillier, France; 2005.
5. Osborne CA, Kruger JM, Lulich J, et al. Feline lower urinary tract diseases. In: Ettinger SJ, Feldman E, eds. *Textbook of Veterinary Internal Medicine*. Philadelphia, PA: WB Saunders; 2000:1710–1747.
6. Kyles AE, Hardie EM, Wooden BG, et al. Clinical, clinicopathologic, radiographic, and ultrasonographic abnormalities in cats with ureteral calculi: 163 cases (1984–2002). *J Am Vet Med Assoc* 2005;226:932–936.
7. Lekcharoensuk C, Osborne CA, Lulich JP, et al. Trends in the frequency of calcium oxalate uroliths in the upper urinary tract of cats. *J Am Anim Hosp Assoc* 2005;41:39–46.
8. Ruby AL, Ling GV. Methods of analysis of canine uroliths. *Vet Clin North Am Small Anim Pract* 1986;16:293–301.
9. Prien EL, Frondel C. Studies in urolithiasis: I. The composition of urinary calculi. *J Urol* 1947;57.
10. Neumann RD, Ruby AL, Ling GV, et al. Ultrastructure and mineral composition of urinary calculi from horses. *Am J Vet Res* 1994;55:1357–1367.
11. Osborne CA, Lulich JP, Thumchai R, et al. Feline urolithiasis: Etiology and pathophysiology. *Vet Clin North Am Small Anim Pract* 1996;26:217–232.
12. Osborne C, Kruger JM, Lulich JP, et al. Disorders of the feline lower urinary tract. In: Osborne CA, Finco D, eds. *Canine and Feline Nephrology and Urology*. Philadelphia, PA: Williams and Wilkins; 1995:625–680.