European Society of Veterinary Clinical Pathology

MYSTERY CASE SESSION

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SPECIMEN: Wright-Giemsa stained, images from smeared cytocentrifuged urine

SIGNALMENT: 15 year-old male castrated Shire horse

HISTORY AND CLINICAL FINDINGS:

The patient presented to the referring veterinarian for a history of chronic urinary incontinence and audible sloshing sounds while trotting. Weight loss was also noted in the previous 6 months. At rest, the owner reported micturition of grossly normal urine with a strong stream. The patient was up to date on vaccinations for tetanus, Eastern and Western Equine Encephalitis, Equine Herpesvirus-1 and Equine Influenza Virus.

The patient was referred to the Atlantic Veterinary College Veterinary Teaching Hospital for further evaluation. On presentation, the patient was alert but mildly restless. Physical examination revealed the patient to be moderately thin with a body condition score of 4/9 and decreased muscle mass over the scapular, gluteal, and dorsal regions. Distal hind limb inflammation with thickened, fissuring skin and multiple sores was noted due to urine scalding. Respiratory rate was moderately elevated at 28 breaths per minute with normal respiratory effort. Other vital parameters were within reference intervals. Normal defecation was observed during examination, but the patient constantly dribbled urine without posturing to urinate. Blood was collected and submitted for a CBC and serum biochemistry panel. The following day, urine was collected via catheterization and was submitted for urinalysis. Polydipsia was supported by observing excess water drinking on admission. Once renal parameters were known, water was rationed rather than provided ad libitum.

LABORATORY DATA:

Hemogram results: Sysmex XT-2000iV Hematology Analyzer

TEST	FLAG	UNITS	RESULT	REFERENCE INTERVAL
RBC		x 10^{12}/L	6.9	6.5 – 12.5
HCT		L/L	0.35	0.32 – 0.52
HGB		g/L	124	110 – 190

MCV		fL	52	34 – 58
MCH		pg	18	11 – 19
MCHC		g/L	350	310 – 370
PLTS		x 10^9/L	314	100 – 600
WBC		x 10^9/L	6.4	5.5 – 12.5
Neutrophils (Seg)		x 10^9/L	4.1	2.7 – 6.7
Bands		x 10^9/L	0.1	0.0 – 0.1
Basophils		x 10^9/L	0.06	0.0 – 0.17
Lymphocytes		x 10^9/L	1.6	1.5 – 5.5
Monocytes		x 10^9/L	0.5	0.0 – 0.8
Toxic change		-	Slight	-
Protein		g/L	76	60 – 77
Fibrinogen		g/L	3	< 5

Biochemistry results: COBAS c501 Biochemistry Analyzer

TEST	FLAG	UNITS	RESULT	REFERENCE INTERVAL
Sodium		mmol/L	138	135 – 148
Potassium		mmol/L	4.2	3.0 – 5.0
Chloride		mmol/L	101	98 – 110
Calcium		mmol/L	3.09	2.8 – 3.44
Phosphorus	L	mmol/L	0.8	1.0 – 1.8
Magnesium		mmol/L	0.83	0.74 – 1.02
Urea	L	mmol/L	2.3	3.5 – 7.0
Creatinine		µmol/L	122	78 – 143
Glucose	H	mmol/L	6.6	3.6 – 5.6
T. Bilirubin		mmol/L	10	4 – 102
ALP		U/L	204	95 – 233
GGT		U/L	12	0 – 25
SDH		U/L	6	1 – 15
AST		U/L	250	197 – 429
CK		U/L	278	50 – 500
T. Protein		g/L	75	60 – 77
Albumin	L	g/L	18	25 – 36
Globulins	H	g/L	57	24 – 44
A:G Ratio	L	-	0.32	0.60 – 1.50

Urinalysis results: Collected via catheterization

MACROSCOPIC TESTS	RESULT
Appearance	Yellow/Cloudy
USG	1.010
Protein	Trace
Glucose	Negative
Ketone	Negative
Blood	Trace
pH	7.5
Sulfosalicylic acid precipitation	1 g/L
Urobilinogen	1.7 µmol/L

MICROSCOPIC TESTS	RESULT
WBC/400x	TNTC
RBC/400x	6 – 10
Epithelial/400x	0
Crystals	++
Casts/100x	0
Bacteria	++++
Fat	–
Debris	–

Images from Wright-Giemsa stained, smeared cytocentrifuged urine dry-mount are provided for evaluation.

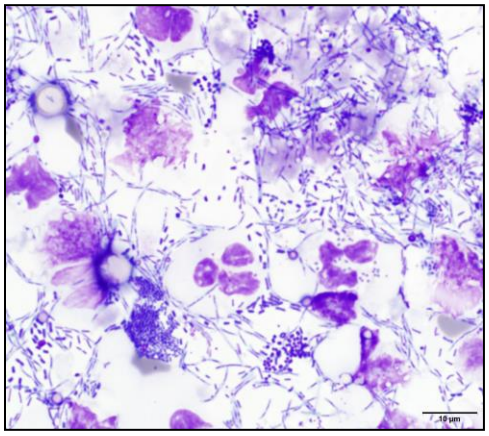

Figure 1. Wright-Giemsa stained, smeared cytocentrifuged urine sediment

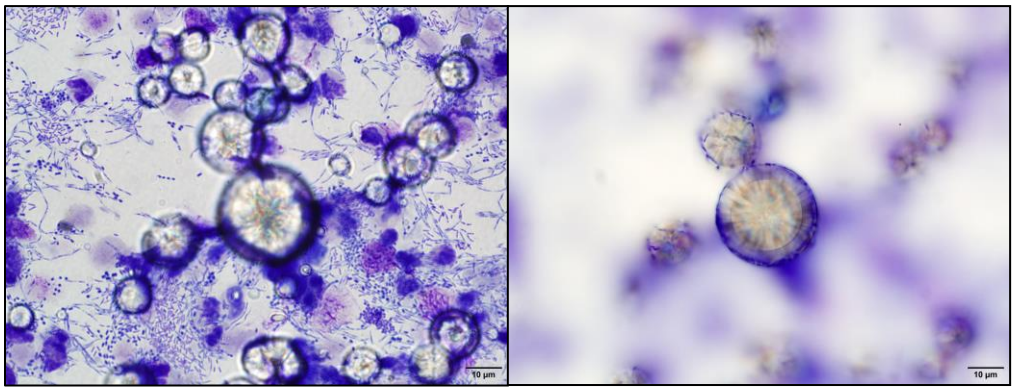

Figure 2. Wright-Giemsa stained, smeared cytocentrifuged urine sediment

ADDITIONAL DIAGNOSTIC TESTS:

Aerobic bacterial culture of urine resulted in multiple strains of *Escherichia coli* and a single *Enterococcus* species. Anaerobic bacterial culture of urine resulted in a single *Peptostreptococcus* species. The patient was unable to fully empty the urinary bladder through normal micturition, necessitating urinary catheterization and manual drainage. Approximately 9 liters of foul-smelling, brown turbid urine was obtained.

QUESTIONS:

1. What is the most likely cause of crystal formation in this urine?
    a. Inappropriately alkalotic urine
    b. Clinically normal finding in horses
    c. Hepatic injury or insufficiency
    d. Artifact precipitation with delayed analysis

2. Which of the following is most likely to cause urinary incontinence as seen in this horse?
    a. Neurologic dysfunction
    b. Urinary obstruction
    c. Urinary tract infection
    d. All of the above

CYTOLOGIC DESCRIPTION:

The smeared cytocentrifuged urine sample is adequately cellular with poor cell preservation on a non-staining to pale blue-lavender background containing large amounts of magenta granular, dendritic to occasionally streaming debris. Nucleated cells consist of markedly degenerate neutrophils present individually and in variably sized dense clumps. Myriads of pleomorphic bacteria are predominantly found throughout the background, but also occasionally in neutrophils. Bacteria comprise cocci, diplococci, rods, and beaded filamentous bacteria (Figure 1). Also present are moderate numbers of tan-brown to occasionally colourless, ~ 1 – 50 um diameter circular, radially striated crystals and infrequent clear small elliptical to polygonal crystals (not depicted in provided images), both of which refract polarized light (Figure 2).

CYTOLOGIC INTERPRETATION/DIAGNOSIS:

Marked septic neutrophilic inflammation
Moderate calcium carbonate crystalluria

ADDITIONAL FINDINGS:

Transrectal/Transabdominal ultrasound revealed bilateral dilatation of renal pelvises and ureters. The urinary bladder wall was thickened and a thick layer of sediment was observed ventrally within the urinary bladder (Figure 3).

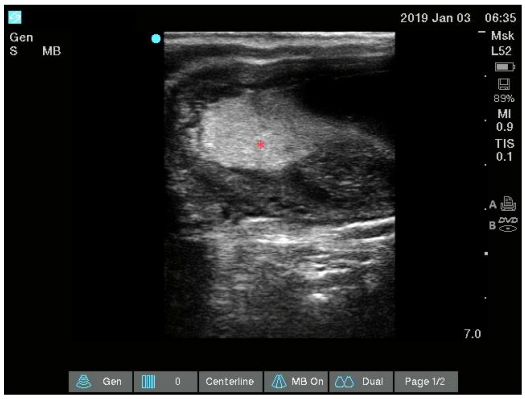

Figure 3. Transrectal ultrasound demonstrating large volume of hyperechoic material present on the ventral surface and free-floating within the urinary bladder (marked with an *).

DIAGNOSIS:

Idiopathic urinary bladder paralysis with sabulous cystitis

CLINICAL OUTCOME/FOLLOW-UP:

The patient was admitted to the Atlantic Veterinary College Veterinary Teaching Hospital and hospitalized for 22 days. During this time the patient's urinary bladder was evaluated via cystoscopy (Figure 4). The urinary bladder was evacuated via catheterization and lavaged with acetylcysteine on six occasions. The volume of urine removed prior to lavage varied, but on

multiple occasions was greater than 15 liters. Although the amount of sediment and debris was significantly reduced with these treatments, incomplete urinary bladder emptying led to continued calcium carbonate sedimentation and prevented complete resolution.

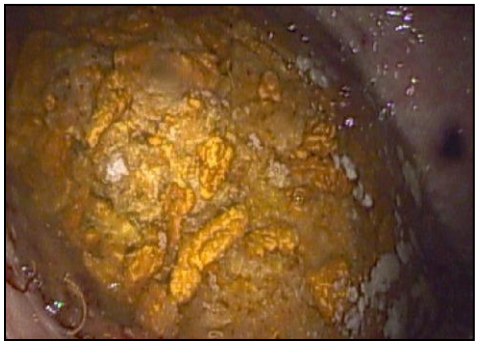

Figure 4. A large quantity of golden-yellow to tan sabulous material identified within the urinary bladder via cystoscopy. Multifocal small areas of hemorrhage are also present along the urinary bladder wall.

The patient was discharged with instructions to continue performing intermittent urinary catheterization and urinary bladder lavage every 3 – 4 weeks indefinitely. Treatments continued intermittently for 2 years, but the patient's health continued to deteriorate throughout that time. Two days prior to planned humane euthanasia, the patient was found dead. A field necropsy was performed and the urinary bladder contained a moderate amount of sabulous material approximately 1 cm thick, but no sediment concretions or uroliths.

ANSWERS TO QUESTIONS:

1. b
2. a

DISCUSSION:

Sabulous cystitis is a term describing an accumulation of inspissated material and associated inflammation within the urinary bladder.[1] This is typically associated with inadequate urinary bladder emptying due to either neurologic deficits or non-neurologic urinary incontinence.[2,3,4] Neurologic deficits can be associated with conditions such as polyneuritis equi (previously *cauda equina neuritis*), Equine Herpesvirus-1 infection, sorghum cystitis-ataxia syndrome or lesions of the lumbosacral spinal cord which lead to lower motor neuron urinary bladder signs, although the majority are classified as idiopathic.[1,2,4] In health, micturition is stimulated when the urinary bladder contains approximately 3 – 4 liters of urine.[4] The lack of active urination and initial drainage of 9 liters of urine from this patient via catheterization indicates significant distention and stretching of the urinary bladder wall. The sloshing sounds reported by the owner while this patient was trotting likely reflected agitation of the increased urine volume with shifting of the sabulous material. These cases generally carry a poor prognosis as at the time of diagnosis most will have irreversible dysfunction of the urethral sphincter and detrusor muscles.[1] These patients require continued care in the form of regularly scheduled urinary bladder lavage, or at minimum urinary catheterization, to remove retained urine and sabulous material.[1] In addition to urinary bladder lavage, it has been previously suggested that feeds high in calcium, such as alfalfa or beet pulp, should be avoided in horses with sabulous cystitis to reduce calcium excretion and hopefully slow sediment accumulation.[1]

The sabulous material consists predominantly of inspissated mucus and calcium carbonate crystals, but can also include components of other crystals such as calcium phosphate, sulfate or oxalate.[5] In health, features such as the high mucus content of equine urine help prevent urolith formation.[4] However, incomplete micturition leads to ventral accumulation of mucus and crystalline material which inspissates with time.[4] This further exacerbates neurologic dysfunction by stretching the urinary bladder wall and resisting detrusor muscle contraction, leading to worsening urinary bladder contractility and continued accumulation of sabulous material.[4] A combination of mucosal irritation from this material and ammoniagenesis by associated bacteria is thought to cause the accompanying cystitis, leading to further damage to the urinary bladder wall and musculature.[1]

Bacterial urinary tract infections are uncommon in horses, but when present are usually secondary to impairments or obstructions which alter the flow of urine.[6] Previously reported etiologic agents include *Escherichia coli*, *Enterobacter* spp, *Proteus* spp, *Klebsiella* spp, *Pseudomonas* spp, and *Corynebacterium* spp.[6] In cases of sabulous cystitis, the accumulated material may provide protection for bacteria similar to a biofilm. This potentially inhibits antimicrobial penetration and efficacy, necessitating physical removal of the sabulous material prior to antimicrobial administration.

In the current case, no causative condition was identified for the urinary bladder paralysis and no other neurologic deficits were identified. The leukon revealed slight toxic change which was attributed to inflammation. No other support for inflammation on the CBC was evident, possibly due to confinement of inflammation to the urinary bladder and distal hind limbs. The mild hypoalbuminemia was suspected to reflect an acute phase inflammatory response. However, given the concurrent mild proteinuria, selective renal loss of albumin contributing to the hypoalbuminemia could not entirely be excluded, although the proteinuria was suspected to be solely post-renal in origin due to the mild hematuria and marked pyuria. The mild hyperglobulinemia was attributed to antigenic stimulation, but the presence of a developing monoclonal gammopathy could not be ruled out. Serum and urine protein electrophoresis were not performed. An underlying cause of the patient's polydipsia was not isolated, but the hypophosphatemia, decreased urea and isosthenuria were attributed to the presumed secondary polyuria. Decreased intake, given the low body condition score, could also have contributed to the hypophosphatemia and low urea. The hyperglycemia was attributed to stress or a physiologic excitement response.

Urinalysis revealed many calcium carbonate crystals, but also fewer clear, small, elliptical to polygonal crystals most consistent with calcium oxalate monohydrate and dihydrate crystals. Clear evidence of septic neutrophilic inflammation was also a prominent finding on urinalysis. Antimicrobial therapy was not initiated due to the large volume of sabulous material present, likelihood of antimicrobial failure and concern for the development of antimicrobial resistance. Frequent urinary bladder lavage to remove the sabulous material, as well as reduce the bacterial load was intended to decrease detrusor muscle stretching, improve antimicrobial efficacy (if antimicrobial use was to be elected) and improve patient comfort. Commonly used for its mucolytic properties while treating endometritis, acetylcysteine was added to the lavage fluid to help break apart the sabulous material and ease urinary bladder evacuation.[7]

Given the infrequency of urinary tract infections in horses, particularly males, it is important to consider all possibilities when septic urine is identified. This includes the presence of sabulous material and underlying neurologic deficits. Sabulous cystitis is an uncommon condition which may not be widely known outside of equine clinician circles. Urinalysis results indicative of a

urinary tract infection, paired with history suggesting urinary dysfunction, should raise suspicion for sabulous cystitis despite the lack of specific urinalysis results to indicate this condition.

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REFERENCES:

1. Rendle DI, Durham AE, Hughes KJ, Lloyd D, Summerhays GES. Long-term management of sabulous cystitis in five horses. Vet. Record. 2008; 162: 783-788.
2. Watson ED, McGorum BC, Keeling N, Clarke CJ, Mayhew IG. Oestrogen-responsive urinary incontinence in two mares. Equine Vet. Educ. 1997; 9(2): 81-84.
3. Keen JA, Pirie RS. Urinary incontinence associated with sabulous urolithiasis: a series of 4 cases. Equine Vet. Educ. 2006; 18(1): 11-19.
4. Schott II HC, Waldridge B, Bayly WM. Disorders of the urinary system. In: Reed SM, Bayly WM, Sellon DC ed. Equine Internal Medicine. 4th ed. St Louis, MO: Elsevier Inc; 2018: 888-990.
5. Diaz-Espiñeira M, Escolar E. Infrared and atomic spectrometry analysis of the mineral composition of a series of equine sabulous material samples and urinary calculi. Research Vet. Sci. 1997; 63: 93-95.
6. Saulez MN, Cebra CK, Heidel JR, Walker RD, Singh R, Bird KE. Encrusted cystitis secondary to *Corynebacterium matruchotii* infection in a horse. JAVMA. 2005; 226(2): 246-248.
7. Schnobrich MR. Disorders of the Reproductive Tract. In: Reed SM, Bayly WM, Sellon DC ed. Equine Internal Medicine. 4th ed. St Louis, MO: Elsevier Inc; 2018: 1217-1364.