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An ABAXIS International Publication

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Words from the Editor of Vetcom Publications

Welcome to Vetcom, an Abaxis International Publication, volume #57. Vetcom is read by veterinarians, veterinary technicians and office staff from all over the world, therefore our issues always have some international news and cases. Vetcom offers readers case studies, practice tips from a clinical perspective as well as educational opportunities and recent news from Abaxis.

DISCOVER THE LATEST AND RE-IMAGINED VETCOM!

TO start, let me say that I am a magazine enthusiast, a junkie who from my earliest school days has been obsessed with flipping through the pages of magazines, first absorbed in their images and stories, later assigning and editing my own. I really believe that no matter whether a magazine is delivered to your doorstep or to your computer, printed on glossy stock or on cheap tabloid paper, appearing on your iPad or your cell-phone screen, it is still and foremost the work of an editorial team for a discerning audience, to provide an educational and impactful variety of case studies and images from an esteemed group of specialists prepared for our readers.

While technology efficiently delivers news stories to our desktops, laptops and mobile devices, magazines are all about context—how ideas and images are presented in relation to one another and within a larger point of view. Magazines are about trust and partnership: We, the editors, will strive always to keep you engaged; you, the readers.

Furthermore, after 10 years of consciously working on improving Vetcom to include cases from around the world, we have again upped the ante and redesigned and reimagined this and all issues of Vetcom going forward. I hope you enjoy it.

Stay with us, and expect more.

Sincerely,

Valerie Goodwin-Adams
Editor-in-Chief - Vetcom Publications
Director, Global Marketing
valeriegoodwin@abaxis.com
510-675-6604

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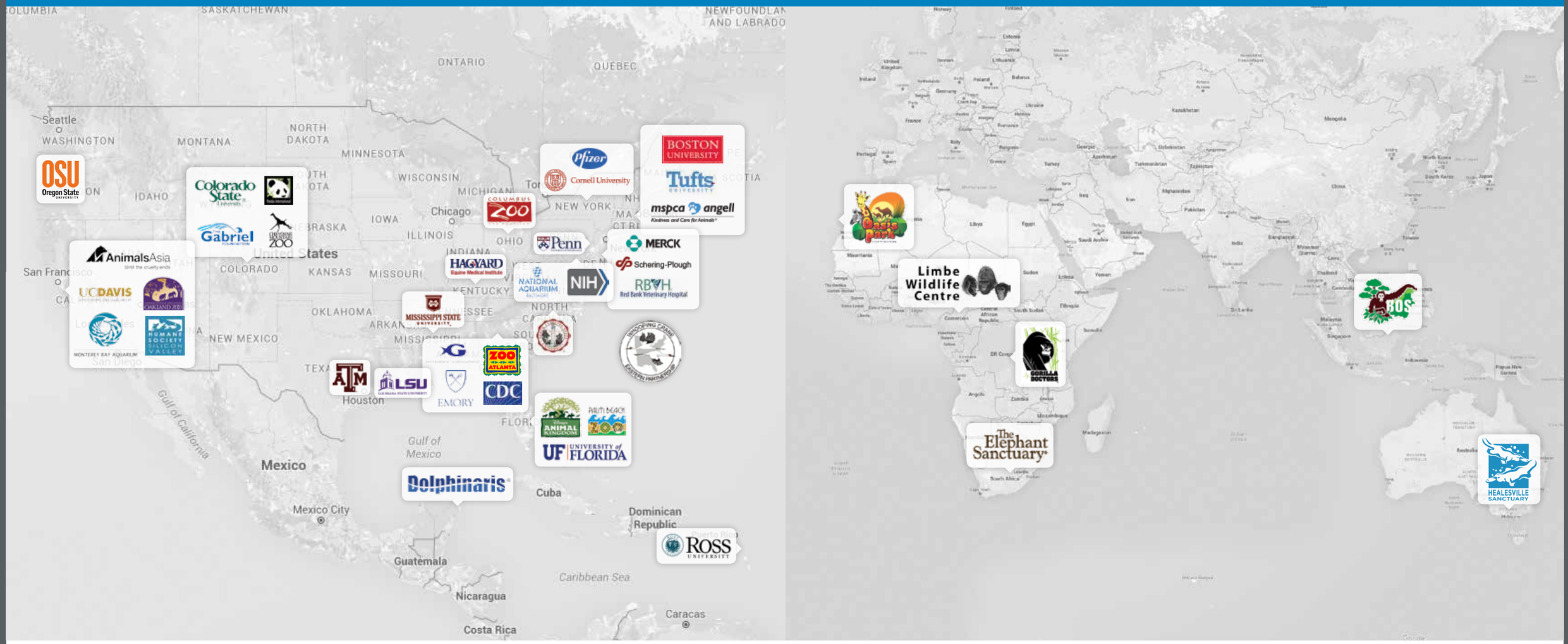

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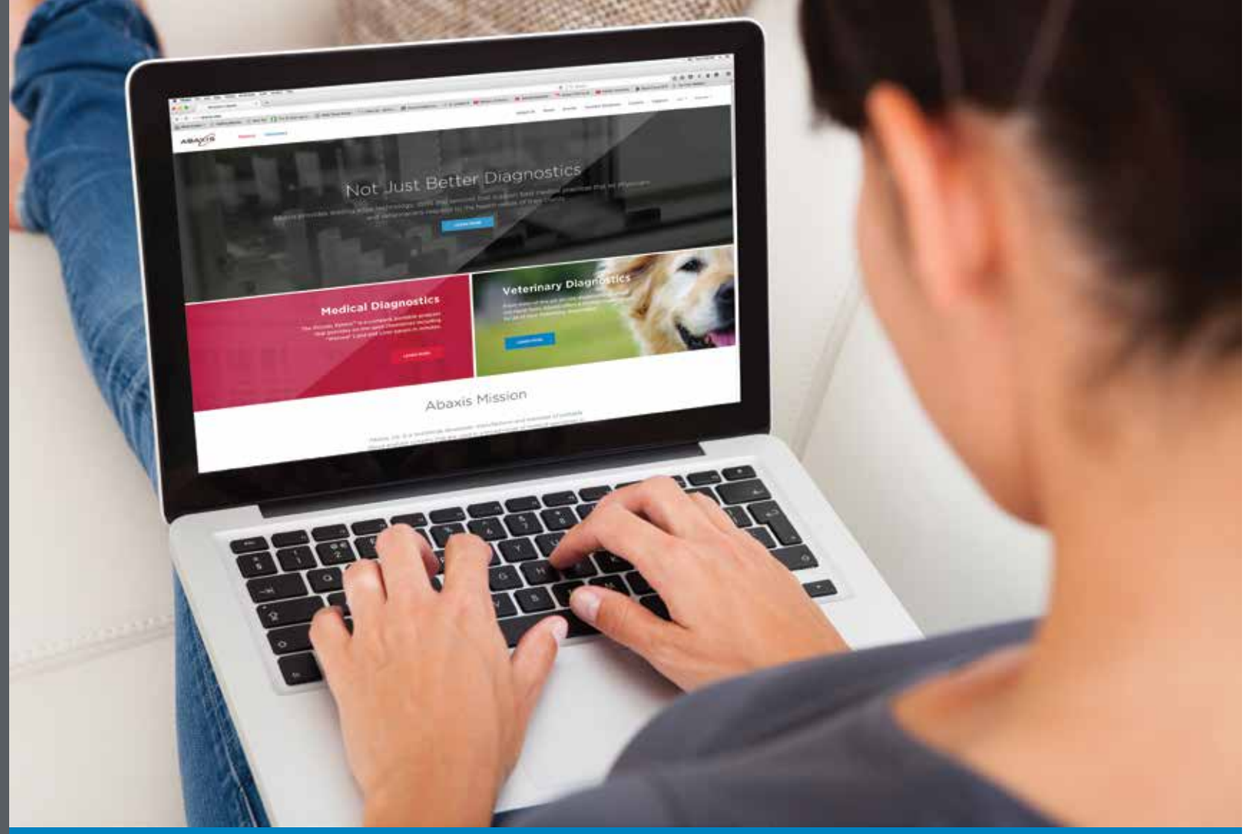
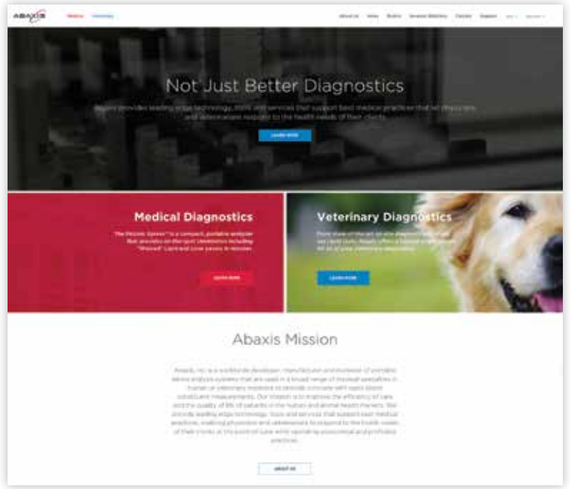


Abaxis Launches

New 5 Language Global Website

THE new Abaxis website is a global website designed to provide a unique experience for markets worldwide. Every page has been customized to guide visitors to only view Abaxis products available in their respective countries. Visitors to the Veterinary section of the site will be able to choose from seven specialties that will further guide them to Abaxis products, stories, and testimonials that best relate to the largest markets but will continuously expand its capabilities as we enter new markets. New features of the website include mobile responsiveness which will enable visitors to view custom sized versions of the website whether they are on a desktop, laptop, tablet, or phone. We have also redesigned the reference center, careers page, and events page. The support center is also brand new to assist our worldwide customer base.



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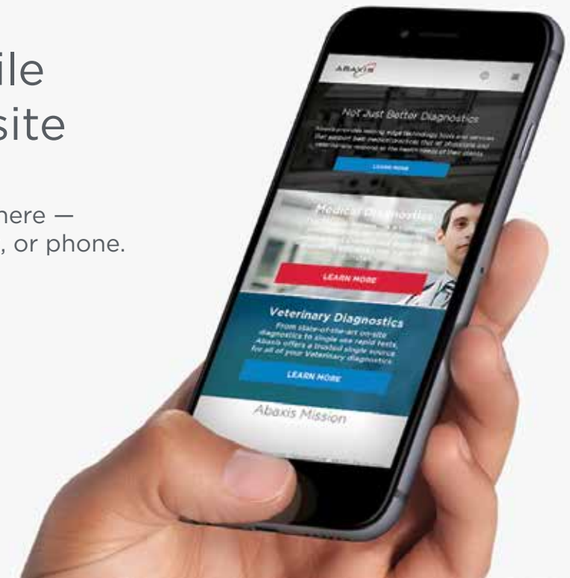


Featured Case

EOSINOPHILIA IN A CAT

Contributing Author:
Heidi Ward, DVM, ACVIM (Oncology)

A 6 year old Male neutered DSH presented with a 1 month history of lethargy, anorexia, vomiting, diarrhea and weight loss. Previous diagnostics included a fecal floatation, FeLV/FIV, CBC/Profile and Heartworm tests. The only abnormality found upon initial work up was a peripheral leukocytosis of 31,500/ul consisting of a mature neutrophilia (12,915/ul), eosinophilia (14,175/ul) and monocytosis (1,260/ul). The cat was subsequently started on therapy with Pyrantel pamoate, Amoxicillin/Clavulanic acid, Fenbendazole and Metronidazole however no improvement was seen and was referred for further diagnostic work up 2 weeks later. On physical examination a palpable abdominal mass was detected. A repeat CBC, profile, thoracic and abdominal radiographs and abdominal ultrasound were performed. Thoracic radiographs were normal; abdominal radiographs revealed an increased opacity within the mid-abdomen and diffuse splenomegaly. The results of the CBC revealed a persistent leukocytosis (35,300/ul) with marked eosinophilia 22,945/ul.

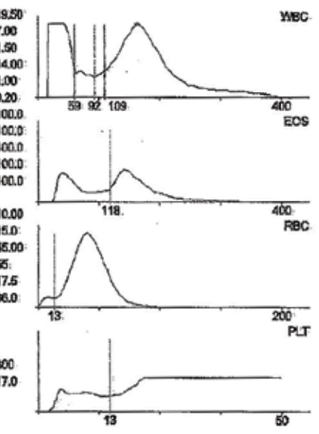
CBC Results

GulfCoast Vet Oncology/Internal Medicine
7519 S Tamiami Trail
Sarasota, FL 34231
(941)926-1920

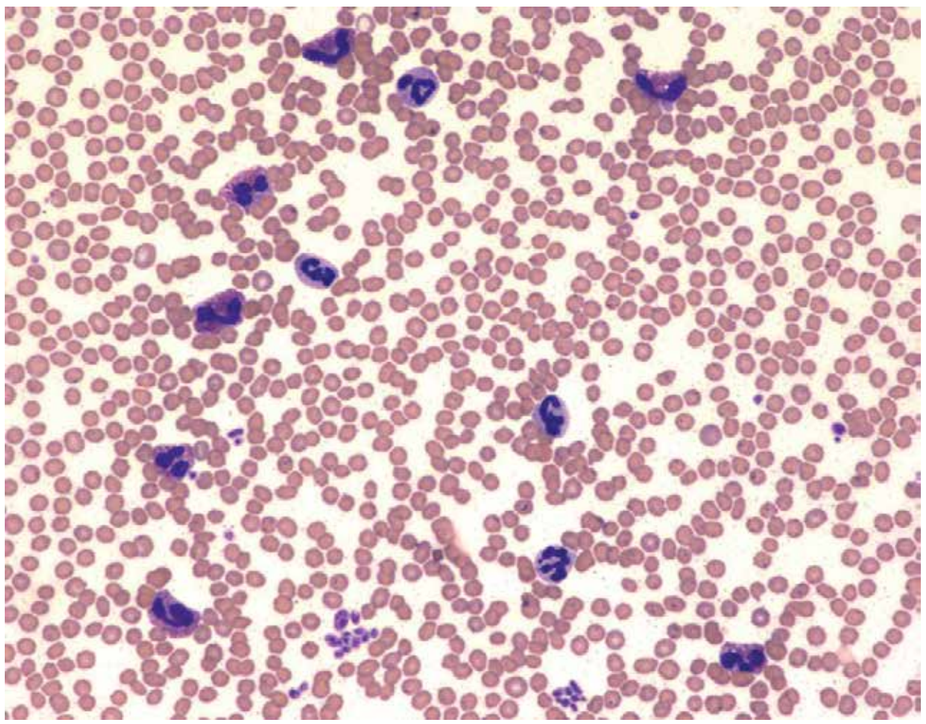
Sample ID	00572	Name	
Patient ID		call	Age 9y
Type		Cat	Gender Spayed
Test date	04/28/2014 08:30 AM	Serial No.	380059
Report date	04/28/2014 08:45 AM		
Doctor	ward		

WBC	65.79+	10^9/l	5.50 - 19.50	
LYM	6.35	10^9/l	1.50 - 7.00	
MON	3.32+	10^9/l	0.00 - 1.50	
NEU	39.00+	10^9/l	2.50 - 14.00	
EOS	16.48+	10^9/l	0.00 - 1.00	
BAS	0.64+	10^9/l	0.00 - 0.20	
LYM%	9.7	%	0.0 - 100.0	
MON%	5.1	%	0.0 - 100.0	
NEU%	59.3	%	0.0 - 100.0	
EOS%	25.0	%	0.0 - 100.0	
BAS%	1.0	%	0.0 - 100.0	
RBC	6.63	10^12/l	5.00 - 10.00	
HGB	9.2	g/dl	8.0 - 15.0	
HCT	26.50	%	24.00 - 45.00	
MCV	40	fl	39 - 55	
MCH	13.9	pg	12.5 - 17.5	
MCHC	34.8	g/dl	30.0 - 36.0	
RDWc	26.0	%		
RDWs	43.0	fl		
PLT	221-	10^9/l	300 - 800	
MPV	11.3-	fl	12.0 - 17.0	
PCT	0.25	%		
PDWc	35.9	%		
PDWs	13.5	fl		

Warning W

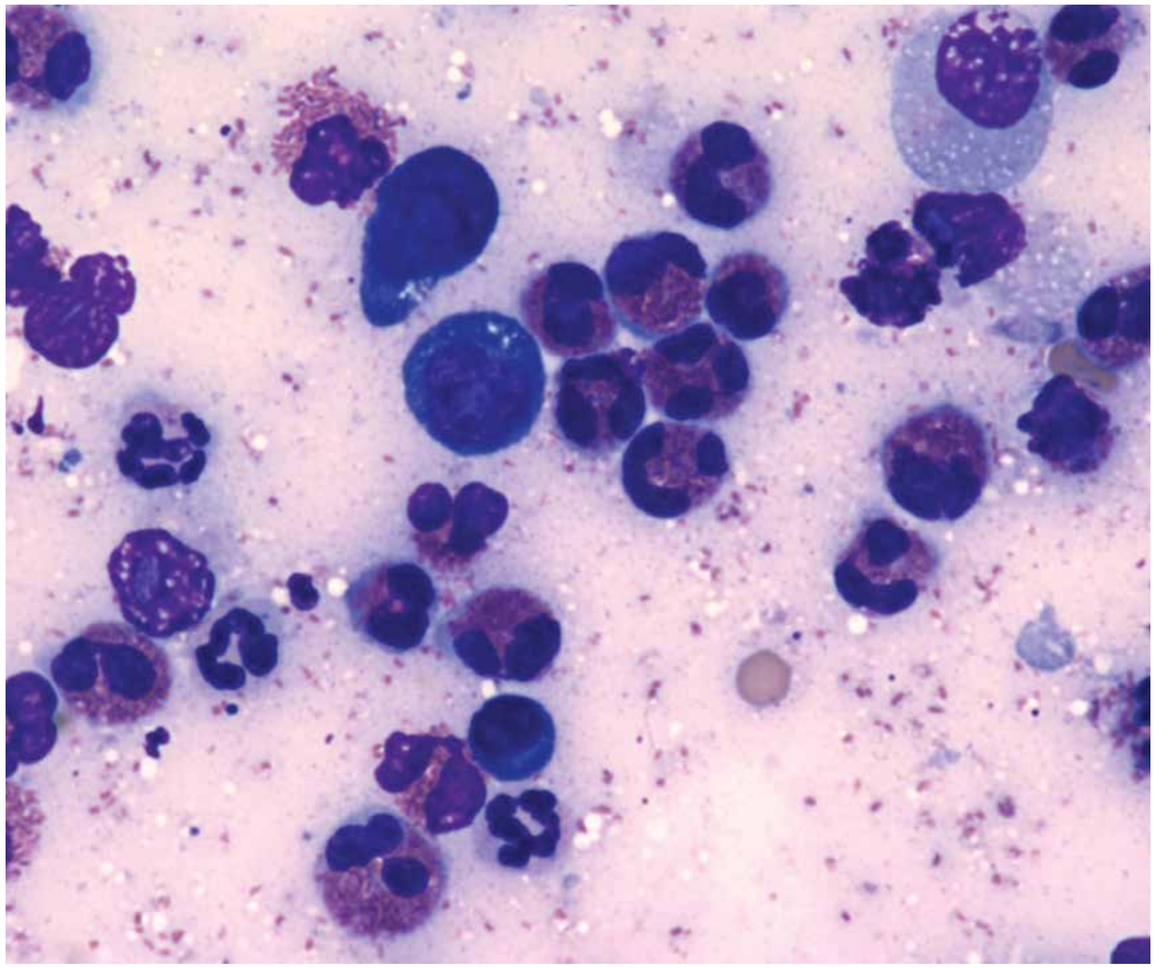


CBC Smear



Marked Peripheral Eosinophilia.

Cytology of Intestinal Mass



Nucleated cells consisted primarily of eosinophils found individually and in groups. Occasional foamy macrophages and a few apical round cells with nuclear diameters reanging from 12 - 15 microns were noted.

On ultrasonography of the abdomen, a 2.7 x 1 cm focal poorly echogenic circumferential small intestinal wall thickening with luminal narrowing was seen. No lymphadenopathy was detected. A fine needle aspirate of the intestinal mass was performed which revealed a predominant population of eosinophils found both individually and in groups. Occasional foamy macrophages and a few atypical round cells with nuclear diameters ranging from 12-15 um were also present. These cells had features consistent with atypical medium lymphocytes (ie round nuclei, coarsely granular chromatin, prominent nucleoli and scant amounts of deeply basophilic cytoplasm). In light of the presence of an eosinophilic inflammatory intestinal mass, atypical lymphocytes and peripheral eosinophilia, a differential diagnoses of hypereosinophilic syndrome or intermediate cell lymphoma with tumor associated eosinophilia was made.

An exploratory laparotomy to determine feasibility of resection of the intestinal mass and to further characterize the atypical cells and rule out underlying neoplasia was declined by the owner. The cat was subsequently given dexamethasone 4 mg SQ and released on Prednisone 5 mg q12 hr. No further vomiting or diarrhea was seen by the owner subsequent to steroids within 48 hours. On recheck 2 weeks later, the cat had gained ½ pound, the eosinophilia had resolved (870/ul) and ultrasonography of the abdomen revealed a reduction in size of the intestinal mass by 75% with no evidence of mesenteric lymphadenopathy or splenomegaly. The cat was continued on Prednisone 5 mg q 12hr. 3 months later the cat represented for vomiting and anorexia. A repeat CBC revealed a nonregenerative anemia and recurrence of marked leukocytosis 65,800/ul with eosinophilia 29,610/ul. Ultrasonography revealed evidence of intestinal thickening, multifocal mesenteric lymphadenopathy and splenomegaly. Further therapy was declined and humane euthanasia was elected.

At necropsy, mesenteric lymphadenopathy and splenomegaly were detected and samples were submitted for histopathology. The lymph node revealed infiltration and replacement of normal architecture with large numbers of eosinophils intermixed with fewer mast cells, lymphocytes, plasma cells and fewer macrophages. There were also broad interconnecting trabeculae of fibrous tissue that contained plump fibroblasts which markedly distorted and expanded the lymph node. These changes have been seen in association with gastrointestinal eosinophilic sclerosing fibroplasia. The spleen revealed that the red pulp was infiltrated by large numbers of well differentiated eosinophils suggestive for hypereosinophilic syndrome.

In cats, eosinophilia is defined as an absolute increase in the number of circulating eosinophils of more than 800/ul of blood.[1] Increases up to 5,000/µL are considered mild, up to 10,000/µL moderate and over 20,000/µL marked. General categories of diseases associated with eosinophilia include parasitic and infectious diseases, hypersensitivity disorders, eosinophilic infiltrative diseases, and neoplasia. In a retrospective study of 312 cats with eosinophilia only cats with flea allergy dermatitis, gastrointestinal disease, focal inflammation/eosinophilic granuloma and miscellaneous dermatopathies had eosinophil counts over 10,000/ul.[2] Paraneoplastic eosinophilia has been reported in association with mammary tumors, leiomyosarcoma, T-cell lymphoma, fibrosarcoma, mast cell tumors and urinary bladder tumors.[3] Hypereosinophilic syndrome (HES) is a disorder characterized by a persistent peripheral eosinophilia and organ infiltration by eosinophils, which eventually cause organ failure and death. It occurs primarily in cats and its origin is unknown.[12,13,14,15] Peripheral eosinophil counts in cats with HES range from 3,500/uL to 130,000/uL and the cells appear mature. The distinction between an eosinophilic leukemia and idiopathic hypereosinophilic syndrome remains elusive. Eosinophilic leukemia is diagnosed when serial evaluation of blood reveals prolonged, persistent marked eosinophilia with immature forms and the bone marrow is dominated by a disordered differentiation and maturation of eosinophils.[16,17]

The organs most commonly infiltrated with eosinophils in HES include the bone marrow, gastrointestinal tract, spleen, mesenteric lymph nodes and liver, but any organ can be affected. In humans, most morbidity and mortality is due to cardiac involvement. Clinical signs of HES are nonspecific and vary with the organ(s) affected and include diarrhea, weight loss, anorexia, pyrexia and pruritus. HES is typically but not always a progressively fatal disease. Management consists of lifelong therapy with corticosteroids.[11] Prednisone is started at 4-6 mg/kg /day for at least one month then gradually tapered to 1-2 mg/kg every 24 to 48 hours. Monitoring consists of repeating CBCs and careful abdominal palpation with imaging.

Causes of Intestinal eosinophilic inflammation include parasites, bacteria, food or environmental antigens, fungi, oomyctes, Toxoplasma gondii and herpesvirus. Eosinophils have been shown to produce numerous mediators such as TGF-B and IL-1b which play a role in fibrosis. A unique form of a feline eosinophilic lesion that appears to be limited to the gastrointestinal tract and regional lymph nodes has been termed Feline Gastrointestinal Eosinophilic Fibroplasia (FGESF).[4,5,6,7,8,9,10] The term FGESF was first proposed and described in 25 cats by Craig et al in 2009.4 In this subset

CONTRIBUTING AUTHORS | FEATURED CASE

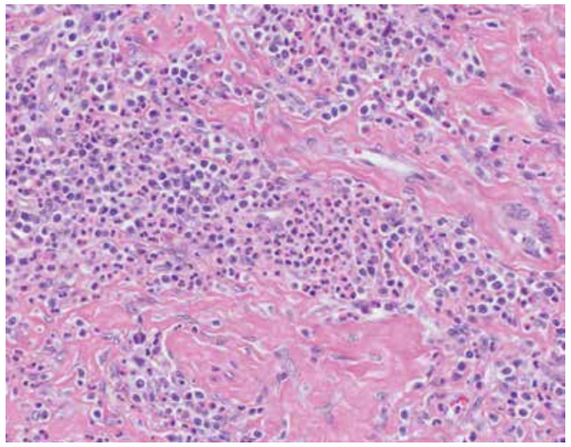
EOSINOPHILIA IN A CAT

of cats, gastrointestinal involvement was most commonly seen at the pyloric sphincter (48%), ileocecocolic junction or colon (36%) and small intestine (16%). A subsequent retrospective detailing 13 cases with similar histologic features has also been reported by Linton et al in 2015.[6] Regional lymphadenopathy was a common finding in both studies. Vomiting, weight loss and a palpable abdominal mass appear to be the most common historical and physical examination findings.

The typical appearance of FGESF is that of an ulcerated mass expanding the wall of the site affected with a characteristic histologic finding of dense collagen trabeculae, fibroblasts and eosinophils. Cytological findings may reveal an eosinophilic matrix, fibroblasts and eosinophils however in some cases only eosinophils are seen.

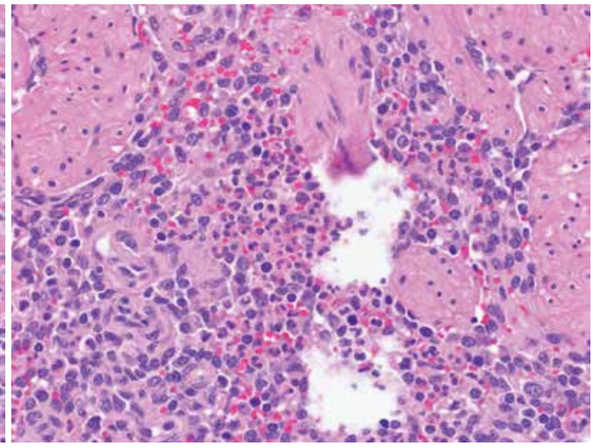
Craig et al postulate that bacteria may be responsible for the initiation of the inflammatory response and were detected in 56% of their cases however antibiotics were not a clinically effective treatment modality. In another study by Ozaki et al 2003, of 27 cats with lesions of similar histologic description bacteria were found in 100% whereas only 69% of cats in the retrospective reported by Linton et al revealed the presence of bacteria. Cats treated with complete excision of their lesions (in cases where feasible) in combination with prednisone had longer survival times than those treated with surgery and antibiotics alone. Prognosis for cats with FGESF is variable and may be associated with the initial location of the lesion as cats with ileocecocolic or colonic masses appear to have longer survivals than those with pyloric involvement as those in the latter group are less surgically accessible to complete excision.[4] In this cat, prednisone alone was helpful in providing an improved quality of life albeit temporarily and therefore steroids alone or in combination with other immunosuppressive agents should be considered in cases in which surgery is not an option.

Lymph Node Histopathology



The lymph node is infiltrated and replaced by large numbers of eosinophils intermixed with fewer mast cells, lymphocytes, plasma cells and fewer macrophages. There were also broad interconnecting trabeculae of fibrous tissue that contain plump fibroblasts which markedly distort and expand the lymph node.

Spleen Histopathology



The red pulp was infiltrated by large numbers of well differentiated eosinophils.

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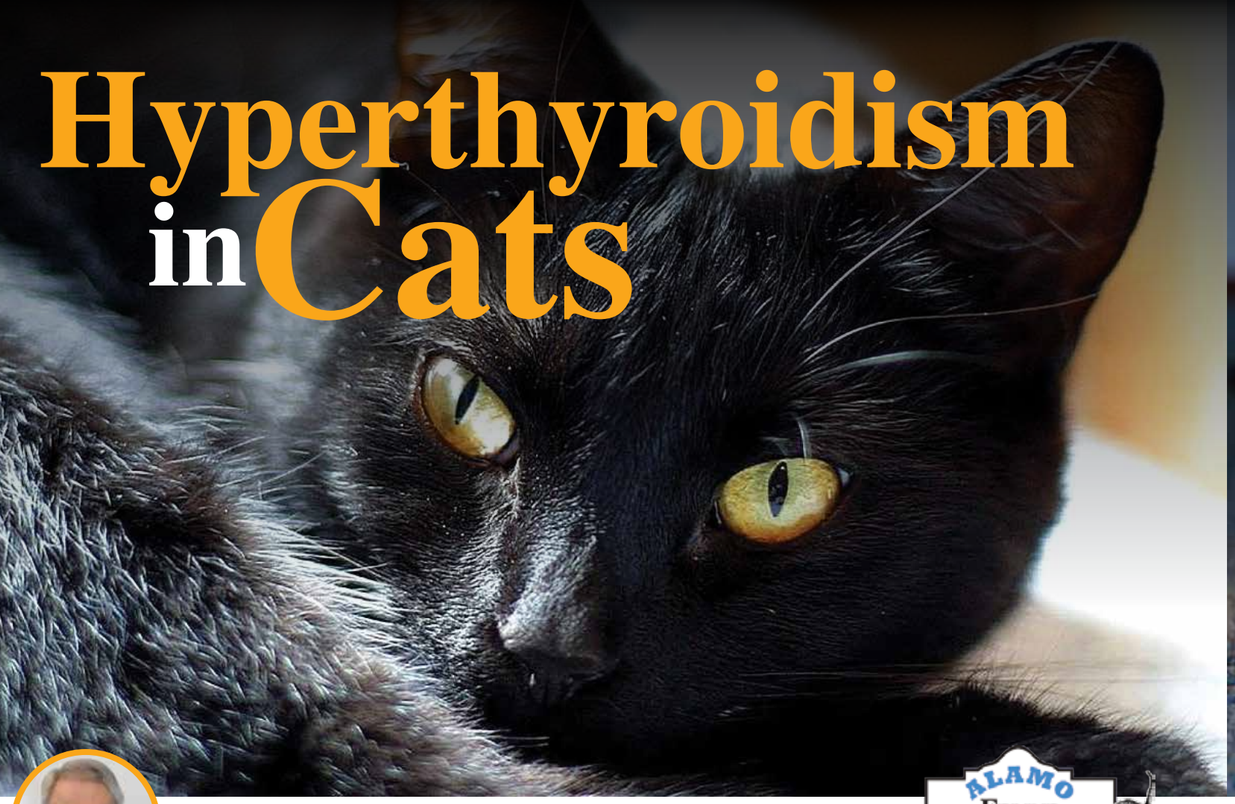
Hyperthyroidism in Cats



Contributing Author:
Gary D. Norsworthy, DVM, DABVP (Feline)
Alamo Feline Health Center | San Antonio, Texas

HYPERTHYROIDISM is the only significant thyroid disease of the cat.

It results from a thyroxine producing tumor that affects one or both thyroid lobes and even ectopic thyroid tissue in the neck or the mediastinum. These tumors begin as adenomas; over time some will progress to adenocarcinomas even if the disease is controlled with methimazole.

The most consistent clinical sign is weight loss due to the accelerated metabolic rate resulting from excessive thyroxine. Weight loss is followed by polyphagia as the cat attempts to compensate; however, the cat cannot eat enough to prevent further weight loss. Weight loss will become severe if the disease is not addressed. Many cats lose well over 50% of their body weight before they are treated or die. Ironically, weight loss is not reported in all hyperthyroid cats. However, this is likely a function of observational failure on the part of the owner. Cats living in households with five or more cats may not be observed carefully. Cats with antisocial personalities are also hard to assess. Cats with long hair coats add further to the difficulty some owners have in detecting early weight loss.
Clinical signs include tachycardia leading to systemic hypertension. This results in increased blood flow through the kidneys increasing glomerular filtration rate (GFR) and decreasing creatinine and blood urea nitrogen (BUN) values. Cats with pre-existing renal disease may be difficult to diagnose due to the artificially lowered creatinine and BUN values.

Hyperthyroidism-induced hypertension may also have adverse, systemic effects. Some of these cats have hypertensive encephalopathy manifested as strokes or seizures. Retinopathy may also occur resulting in retinal hemorrhage, hyphema, and retinal detachment.

The typical age for the onset of hyperthyroidism is about 10 years. However, cats several years younger than this have been well documented.

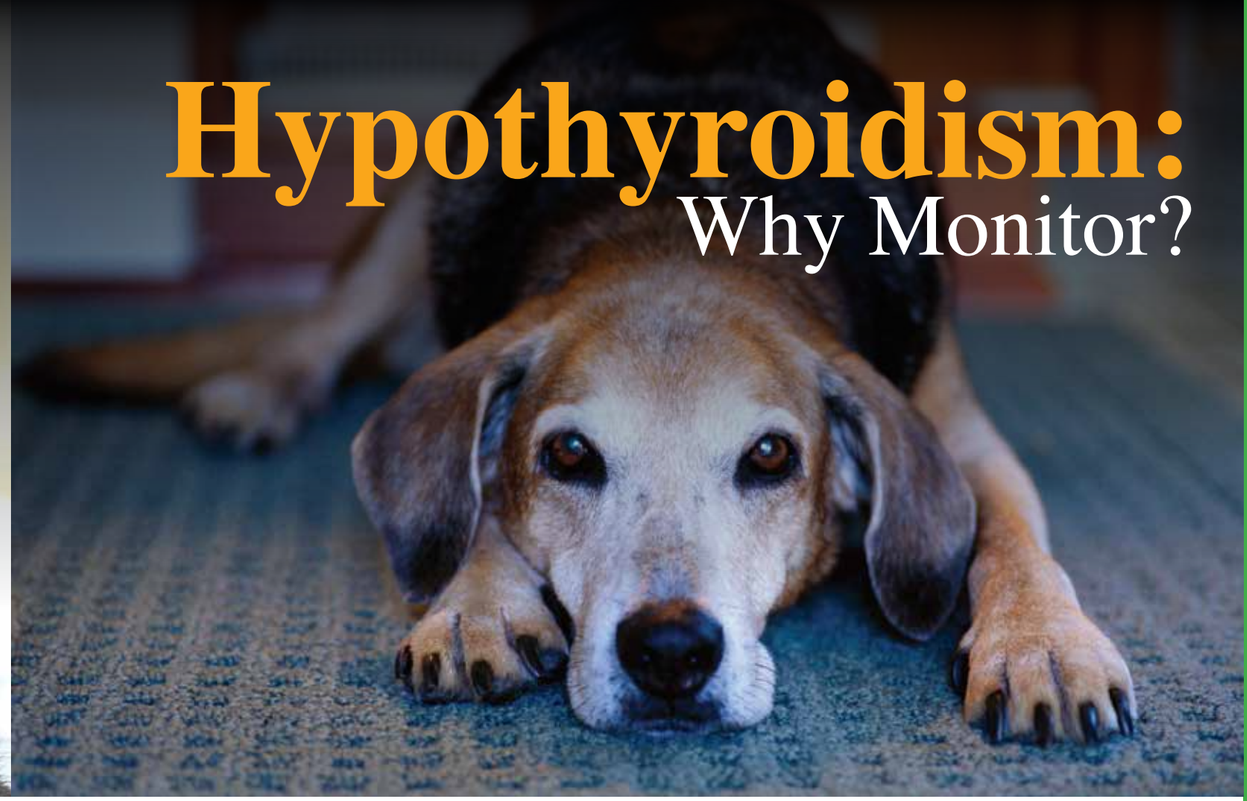
Treatment options include control measures (diet [Hill's y/d] and methimazole) and cures (thyroidectomy and radioiodine therapy). Cure is the most desirable; however, there are many patient and owner factors that are involved in making that choice.

Early detection of hyperthyroidism is highly desirable. Doing so when the total T4 is less than 8 mg/dL usually means that the cat's weight loss has been mild and that tachycardia and hypertension have not yet developed. **The total T4 is our best screening tool.** It is elevated in over 95% of hyperthyroid cats. The total T3 is less sensitive, and the free T4 can be too sensitive, being elevated due to some non-thyroid disease in euthyroid cats. Thyroid stimulating hormone levels have not been shown to be of significant value. If the diagnosis is in question, a T3 suppression test is recommended.

Total T4 screening is highly recommended for all cats over eight years of age and for any cat with unexplained weight loss.

Our goal should be to find hyperthyroid cats in the early stages of disease or even before the onset of clinical signs. Remember that weight loss can elude detection in many cats. Cats diagnosed early suffer no or minimal ill effects from the disease and are very successfully treated If these patients are successfully treated, there is very little chance that their adenoma has progressed to an adenocarcinoma. Early diagnosis greatly increases the chances of returning these cats to a euthyroid, healthy state, giving them many extra years of quality life.

Hypothyroidism: Why Monitor?



Contributing Author:
Tom Lewis, DVM, ACVD

HYPOTHYROIDISM can be a slow but progressive endocrine disorder.

Primary hypothyroidism is the most common type of hypothyroidism in the dog, and is the result of either lymphocytic thyroiditis and idiopathic atrophy of the thyroid gland. Lymphocytic thyroiditis is known to be a progressive disorder, with 80% of thyroid tissue destroyed before clinical signs are seen. The condition may progress over 1-3 years-time and the end results may be the cause of some cases of idiopathic atrophy of the thyroid gland.

Having baseline thyroid levels may allow the clinician to spot a downward trend, and enable the practitioner to institute replacement therapy before serious dermatological or metabolic abnormalities develop.

Extra consideration for annual monitoring of thyroid levels should be given in middle age and older dogs of breeds known to be at risk for the developmentof hypothyroidism. These breeds include (but are not limited to) Golden and Labrador Retrievers and Doberman Pinchers.

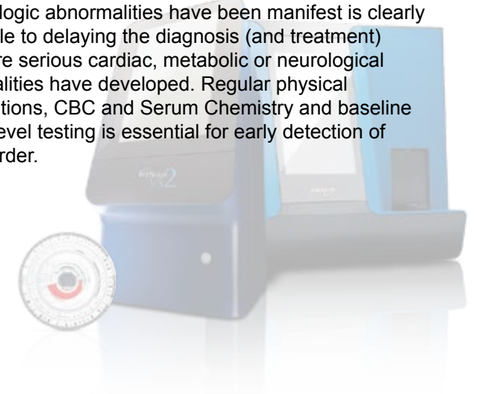
Although measurement of T4 is a good screening test for euthyroidism or hypothyroidism, measurement of combined thyroid function tests including fT4 and TSH is preferred when confirming the diagnosis. The diagnosis can be challenging due to the progressive nature of the disease, conflicting or discordant test results, overlap of T4 levels between euthyroid and hypothyroid patients and variation of normal thyroid levels between different breeds. Further it should be noted that different analyzers and labs have different testing modalities and that each thyroid evaluation has different reference ranges for normal thyroid levels.

An index of suspicion of internal hypothyroidism should be based on appropriate clinical signs and clinical pathology. It is also important to remember certain drugs that will interfere with testing of thyroid levels (T4 and free T4) include glucocorticoids, sulfonamides, NSAIDs, phenobarbital and clomipramine.

Additional clinical pathology changes that should be monitored which provides the clinician a higher degree of suspicion regarding hypothyroidism. These diagnostics include a CBC which may reveal a normocytic, normochromic, nonregenerative anemia and serum chemistries where hypercholesterolemia and hypertriglyceridemia may be seen. Myopathy caused by hypothyroidism may show increases in LDH, ALT, AST and Alkaline Phosphatate.

Some of the early clinical changes of hypothyroidism include lethargy and weight gain, facial myxedema, "cool" skin which may appear to have "bleached out" hairs due to slower turnover. Bilateral alopecia, including sites of wear is also a feature.

Diagnosing a patient with hypothyroidism early when only dermatologic abnormalities have been manifest is clearly preferable to delaying the diagnosis (and treatment) until more serious cardiac, metabolic or neurological abnormalities have developed. Regular physical examinations, CBC and Serum Chemistry and baseline thyroid level testing is essential for early detection of this disorder.



Persistent Polycythemia & Exercise Intolerance in a Horse

Contributing Authors:
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Scott E. McAllister, VMD, Centre Hall Pa.

A 13 year-old Rocky Mountain Horse gelding was initially examined because of lethargy and intermittent loss of appetite. The initial clinical examination and repeated examinations over a six month period were relatively unremarkable except for consistently finding a darker than normal mucous membrane color. (See Figure 1)

Figure 1

Color of oral mucus membranes at hospital admission; HCT of the horse at this time was 53%.

Laboratory testing during this time revealed a persistently high HCT (51-60.9%) with normal MCV, MCH, and MCHC values. Laboratory testing for hepatic or renal dysfunction was uniformly normal. No significant abnormalities were noted in white blood cell counts during this time, although variable elevations in plasma fibrinogen (500-1600mg/dl) using the heat precipitation test and mild increases in serum globulins (4.2-5.2 g/l) were noted. A peritoneal tap was performed and was normal. Courses of treatment with penicillin, oxytetracycline, and trimethoprim-sulfa had minimal effect on the exercise intolerance, but the fibrinogen decreased and body weight improved. Because of the elevation in fibrinogen and persistent polycythemia, a two-week course of treatment with dexamethasone was administered but no clinical change was noted. The horse had gained weight (875 to 950 lbs.) during this time based on girth tape measurements.

The clinical condition did not significantly change over a 6 month time, the persistent elevation in HCT without clinical dehydration suggested a persistent polycythemia (absolute erythrocytosis) and the horse was referred for further examination.

On hospital admission, the temperature was 99.7°F, heart rate 44 bpm, and respiratory rate 24 bpm. The horse appeared bright and his body weight was 1,051 pounds. Normal lung sounds could be heard but were subjectively diminished ventrally. Heart sounds were normal. The hemogram revealed erythrocytosis (HCT 53%), increased RBC (10.8 x 106 µl), but normal MCV (49 Fl), MCH (17 pg), and MCHC (35 g/dl). The leukogram and plasma fibrinogen were normal. A complete serum chemistry panel was normal. Ultrasound of the chest revealed a large amount of hypoechoic fluid in both left and right pleural spaces. Approximately 14 liters of fluid was drained from the chest by placing a chest tube placed in the right hemithorax; repeat ultrasound examination showed that both sides of the thorax had been drained by the right-sided chest tube. Although a mesothelioma was suspected based upon the large volume of pleural transudate (Table 1), mesothelioma could not be confirmed by cytologic examination.

Figure 2

Sonogram of left thoracic cavity of the horse which revealed a large volume of relatively hypoechoic fluid with possible atelectasis of lung (most severe ventral).

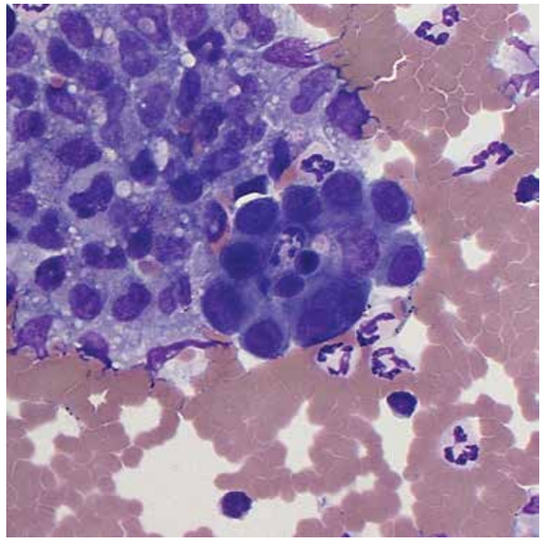
Radiographs and pleuroscopy were offered as additional testing to further confirm the diagnosis but were declined and the horse was discharged. Follow-up examination on the farm two months later revealed an improved attitude, appetite, less exercise intolerance, and a weight gain of 15 pounds although the HCT was 50% and thoracic auscultation and percussion suggested pleural fluid accumulation. A thoracocentesis was performed on the farm and successfully drained a large volume of pleural fluid; the cytologic findings on the fluid were similar to the initial evaluation two months earlier.

The horse appeared clinically normal for several months, even jumping a 3-4 foot fence and HCT decreased into the high 40s.

Fifteen and 18 months later, because of a decline in exercise tolerance; thoracenteses were again performed with drainage of a large volume of pleural fluid (16 and 28 liters) each time; cytologic exam on the last fluid collected had a very low WBC count and no neoplastic cells were seen. Clinical improvement was noted each time after fluid was removed. The horse died one month following the last pleural drainage, 27 months after the onset of clinical signs. Necropsy confirmed pleural mesothelioma, mostly involving the anterior portion of the thorax.

Mesothelioma is a primary neoplasia of mesothelial cells lining the pleural, peritoneal, or pericardial cavities. Unlike in humans where there are known predisposing risk factors, no risk factors have been identified in horses. Although the neoplasia is not common, the clinical feature of large volume body cavity effusion is characteristic. Horses with mesotheliomas have ranged in age from 2-27 years of age. Most commonly, the affected body cavity surfaces are lined by multiple small tumors and effusion, similar to that in the current case report, is often blood-tinged, and has elevated protein with normal WBC, containing various mixtures of non-degenerate neutrophils, lymphocytes, and mesothelial cells. Exfoliation of neoplastic cells is variable with some cases having large sheets of mesothelial cells containing abnormal nuclei and nucleoli while other cases seem to exfoliate poorly. Differentiation between reactive mesothelial cells and neoplastic mesothelial cells can be difficult (See Figure 3).

Figure 3



Cytospin smear (50X) of the pleural fluid from the horse consisted of erythrocytes with moderate leukocytes and several individual and a few clusters of mesothelial cells. Neutrophils are observed within the mesothelial cell clusters and the latter cells are uniform in appearance. The mesothelial cells are likely reactive, but a mesothelioma cannot be ruled out. Photo courtesy of Dr. Ashleigh Newman, Cornell University.

Although multiple cytologic examinations of fluid in this case did not provide a diagnosis, the clinical findings, massive bilateral pleural effusion, and fluid evaluation ruling out infectious or other neoplastic disorders was sufficient to tentatively diagnose pleural mesothelioma. Mesothelioma invasion into other organs or other body cavities is rare; although in the chest both pleural and pericardial membranes may be simultaneously affected.

Horses with pleural mesotheliomas generally present for increased respiratory rate, exercise intolerance and, less commonly, with sternal edema. This case of pleural mesothelioma was of particular interest for several reasons including a prolonged life span with a relatively good quality of life after onset of illness, inability to confirm the diagnosis on repeated pleural fluid cytologic examinations, and persistent polycythemia, all of which are unusual. The persistent polycythemia, which tended to decline slightly following pleural drainage, could be attributed to chronic hypoxia and increased erythropoietin. Measurement of PaO2 prior to draining the chest could have been performed in hospital or on the farm with point of care testing and would have been of interest in this case; a PaO2 value of <80 mm Hg would have been supportive of chronic hypoxia as a cause of the polycythemia. Polycythemia caused by chronic hypoxia in horses is rare. An alternative explanation for the polycythemia would be neoplastic production of an erythropoietin-like hormone or up-regulation of erythropoietin receptors.

The marked increase in fibrinogen, an acute phase protein, which is increased during inflammation, and low plasma protein to fibrinogen ratio were supportive of an inflammatory disease, in spite of a normal WBC. The marked variations in the fibrinogen values during the disease may have been caused by change in inflammation at different time points or differences in methodology, especially with heat precipitation methods.

Differential diagnosis in this case included obstructed or ruptured thoracic duct or other neoplasia. Although lipid or lipid-containing cells were not seen cytologically and both triglyceride and cholesterol levels were low in the fluid, the absence of progression of clinical signs for a prolonged time (>2 years) made thoracic duct disease a consideration.

Table 1: Pleural Fluid Analysis

Date	Color	Total Protein	WBC (µl)	PMNs (%)	Lymphocytes (%)	Macrophages (%)	Comments
3/13	Light Red	5.2 g/dl	1,800	23*	28	49	Transudate effusion, hemorrhage, reactive mesothelial cells
5/13	Yellow	5.4	2,900	47*	25	28	No neoplastic cells identified
8/14	Clear	4.6	1,400	28*	52	18	High protein transudate with evidence of erythrocyte diapedesis; triglycerides (19 mg/dl), cholesterol (49 mg/dl)
11/28	Light Red	4.4	897	-	-	-	Hemorrhage with few scattered inflammatory cells, no neoplastic cells noted.

*Non-degenerate

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URINE MARKING & AGGRESSION: IS IT BEHAVIORAL OR IS IT MEDICAL

Contributing Author:
Debra F. Horwitz, DVM, DACVB

Statement of the problem:

An older cat was presented to the veterinary behaviorist for ongoing urine marking within the home, weight loss and increased aggressive behavior toward people and other cats.

Signalment

The patient was an 11 year old neutered male Ragdoll/Manx mix weighing 13 lbs. at the time of behavioral consultation.

Medical History

The cat was obtained from a shelter in November 1995 and was approximately 8-10 weeks of age. Early medical history from owner noted past urinary blockages which have resolved with dietary intervention and are no longer an issue. For most of the past few years the patient had been overweight (16 lbs.) and fed W/D diet (Hill's Pet Food) without appreciable weight loss until recently. At the most recent wellness visit the patient weighed 14 lbs. and records show the first recorded mention of behavioral issues. The patient had become increasingly aggressive, moody and was barbering his hair and displaying peruria outside of the litter box. Dental disease was also noted and a dental cleaning recommend. Blood work, urinalysis and EKG were performed and all results were WNL. The patient was scheduled for a dental cleaning from which it recovered uneventfully. The cat was also placed in Buspirone 2.5 mg BID to attempt to control the behavioral issues mentioned above. Over the next several months the dosage was increased first to 5 mg BID then 7.5 mg BID. Six months later weight loss, aggression and urine marking were still ongoing. A referral to a veterinary behaviorist was recommended but declined at that time.

The owner then went to another veterinary hospital which was more holistically oriented. In the interim the owner had worked with a trainer and an animal communicator and utilized rescue remedy and Feliway® without any change in the behavior. Treatment with Buspirone was ongoing (5mg TID) at this time, but the behavior had not changed appreciably. The aggressive behavior had escalated with the cat screaming and attacking visitors to the home. The physical examination by the new veterinarian noted hair loss but no other significant physical findings. The veterinarian suggested blood work with a focus on thyroid testing and urinalysis. The patient was sent home with a feline anxiety and stress product. Results of laboratory testing indicated a mildly low WBC, neutropenia, and lymphopenia. Thyroid testing was not particularly diagnostic for hyperthyroidism but the veterinarian was concerned that the weight loss and behavioral changes perhaps signaled an occult hyperthyroidism. Her other diagnostic rule out was a brain lesion of some sort yet undiscovered. The client was dispensed a natural product for early hyperthyroidism (early Hyper-thyrin) to administer daily.

One month follow up showed no change in the patient's behavior the urination continues and aggression had increased. The owner requested treatment with Ovaban which the veterinarian declined to provide and instead the pet was put on Alpha-stim therapy. At this time the owner had already scheduled an appointment with the veterinary behaviorist and agreed to utilize this therapy until that behavior appointment.

Behavioral History

At the time of the behavioral consultation the cat was still on Buspirone 5 mg once daily Feline anxiety and stress and early hyper-thyrin. The behavior had not appreciably changed with these interventions. It was noted that the cat has lost three pounds in the past eight months and currently eats canned Hill's W/D and Natural Balance canned and dry. Two adults and one other cat (11yr. spayed female) were shared the home with the patient. Due to his current aggressive behavior and urine spraying, the patient lived in the basement and only out in the house while on a harness and leash. The cat will also get very agitated at the sight of other cats outside the house. Evidence of over-grooming with subsequent hair loss was noted on his forelegs and side of the body.

Further questioning revealed that the aggressive behavior had actually been present for nearly a year with the patient jumping aggressively at visitors and biting the owner when she attempted to pick him up and aggression toward workmen who come to the home which had not occurred previously. The patient also stalks and attacks the other cat in the home several times a week.

The urination outside of the litter box and possible urine spraying have been present for 10 months or longer. Urine spraying has occurred all over the house including the owner's computer in her home office. The patient continues to utilize a litter box to empty his bladder and bowel. The owners are unclear if the patient is spraying or urinating but related that "They can tell when he is going to do it because he lifts and flutters his tail".

The patient has his owner litter box where he spends his time and it is scooped daily but not emptied, washed and refilled.

Physical findings and examination room behavior

The cat was active and slightly agitated throughout the consultation. He roamed the examination room and investigated everything. His physical condition was normal, although he has lost weight he was not too thin. However, it was noted that his facial features had changed and he looked "jowly" like a mature intact male cat. During the course of the consultation, the patient turned, lifted his tail and urine marked the cabinet. The urine odor was strong and had the distinct smell of an intact tom cat. The owner verified that this is how the urine has always smelled at home.

Diagnosis

Cats with certain medical disorders may display unusual behaviors that can be misinterpreted as a primary behavioral disorder. On the contrary, cats with behavioral issues often must undergo extensive diagnostic testing to rule-out medical disorders before a definitive diagnosis of primary behavioral disorder can be established. Clinical signs such as inappropriate urination, periuria, defecation outside the litter box, eating disorders, poor grooming habits and aggression may be observed in cats with both primary behavioral disorders and primary medical conditions involving the endocrine, gastrointestinal and urinary systems. Two common endocrine disorders of senior cats could account for the symptoms noted in this patient.

Hyperthyroidism is a common feline endocrine disorder that will cause dramatic physical and behavioral change in cats. Hyperthyroidism is quite common in companion cats and may cause hyperphagia, polydypsia, polyuria and loose stools. Cats with endocrine disorders frequently manifest behavioral changes and clinical signs that can be confused with primary behavioral disorders. Cats with clinical signs including aggression, withdrawn interaction with owner, weight loss or gain, poor grooming habits and inappropriate urination should be evaluated for a variety of feline endocrine disorders.

Diagnosis (continued)

The diagnosis is often straightforward based on clinical signs (polyphagia, PU/PD, weight and body condition loss, unkempt hair coat appearance), palpable cervical thyroid gland enlargement and an elevated serum thyroxine (T4) level. However, the diagnosis can be challenging in some cats with appropriate clinical signs, suggestive ancillary physical and laboratory abnormalities that do not have a substantial increase in their T4 value. Approximately 10% of hyperthyroid cats have normal T4 values. Other cats may have early hyperthyroidism or concurrent non-thyroidal disease. Measuring serum free T4 (fT4) can be useful in evaluating cats that are suspected to be hyperthyroid but have normal serum T4 value. Although the sensitivity of fT4 is excellent (98.5%) and fT4 values are less affected by non-thyroidal factors there is an issue with elevated fT4 values in euthyroid cats with other illnesses (i.e., CKD) causing a lower specificity (false positives). The T3 suppression test has been described in cats and can be considered in inconclusive cases. Radioactive thyroid scintigraphy can also be useful in unclear cases as it can identify foci of hyper functional thyroid tissue.

Disorders of the adrenal gland may result in alterations of stress hormones or testosterone, both of which may affect behavior. Increases in testosterone may result in intact male behaviors (urine marking, aggression, mounting) in previously neutered animals and laboratory testing for blood testosterone levels will aid in diagnosis of these disorders. Spontaneous hyperadrenocorticism (HAC) is a less common feline endocrine disorder. Iatrogenic HAC associated with chronic corticosteroid administration or exposure can also been seen in cats. Clinical signs of PU/PD, weight gain, dramatic symmetrical truncal hair loss and epithelial thinning and fragility are seen once the disease is well established. Concurrent DM is present once HAC is established. However, early in the development of HAC the signs are not as dramatic but owners may become aware of abnormal behaviors associated with elevated cortisol levels. If spontaneous HAC is suspected then appropriate testing is indicated. The low dose dexamethasone test is preferred in cats. The dexamethasone dose in cats is 10x higher than dogs as 0.1 mg/kg dexamethasone is used. Serum cortisol is evaluated at 0,4 & 8 hours. An oral dexamethasone suppression test using home-collected urine samples for urine cortisol: creatinine determination has also been described.

Elevated sex hormones can also cause simultaneous medical and behavior signs in cats. Unknown neuter history, bilateral cryptorchid tissue or failure to remove all ovarian or testicular tissue during routine neutering can result in this development. Adrenal gland tumors producing excessive testosterone or estrogen can develop in neutered cats. The increased levels of these hormone will produce typical hormonal male or female behaviors that have been absent in these patients following neutering. Inappropriate urination, urine marking, aggressive behavior, reluctance to interact with owners, hair coat change, etc. are often reported by owners. Ingestion by licking of exogenous testosterone or estrogen agents (medically prescribed for their owners) will cause a similar increase in these hormone and the associated medical and behavioral manifestations. Diagnosis can be confirmed by measuring serum testosterone or estrogen levels. If an elevated hormone level is confirmed and there is no reported exogenous exposure then adrenal gland ultrasound is indicated to determine if a tumor is present.

Urine marking in cats is categorized as either sexual or reactional marking behavior and usually occurs with the cat lifting their tail, backing up to an object, treading with their rear feet and expelling urine backwards onto that object. Cats will mark with urine to attract mates and also urine mark in response to environmental changes and/or stress. Urine marks are often found in socially significant places such as owner possessions, laundry or in prominent locations. Cats that mark with urine on vertical surfaces usually continue to use the litter box for elimination of both urine and stool. The diagnosis should focus on the location of the urine, the size of the urine spot and possible sources of stress in the household. Although spraying is usually thought to be associated with intact animals, neutered animals will spray and cats will often mark inside the house as a territorial response to the presence of outside cats.

Previous laboratory testing and lack of response to intervention made hyperthyroidism unlikely in this patient but should be repeated. The behavior and physical changes would be consistent with additional testosterone either from a retained testicle or an adrenal tumor. If the problem was due to a retained testicle, these behaviors would have likely been noted earlier in this patient's life since he has been in the same home for his entire lifetime. Additionally the tom cat urine odor exuded by the urine sprayed the exam room, make excess testosterone a likely causative agent in the behavior changes.

Behavioral diagnosis were: urine marking secondarily to increased testosterone levels, aggression due to hormonal influences, territorial aggression and underlying anxiety.

Treatment

After the patient was seen for the behavioral consultation, blood was drawn and sent to Michigan State University Diagnostic services. According to the results (p. 29), thyroid values were still within normal ranges but the blood testosterone was elevated for a castrated cat.

Based on the laboratory findings and the sudden behavioral changes an adrenal tumor was suspected. The patient was referred for an ultrasound which confirmed an adrenal tumor. Successful surgery for the adrenal tumor was performed.

The owner was also sent home with standard treatment recommendations for urine marking, litter box maintenance and interactions with visitors.

Urine marking

Previous research has shown that even urine marking cats prefer larger and cleaner litter boxes. The owner was instructed to obtain a larger litter box, continue to scoop out the litter box daily, empty wash and refill it every 7-10 days.

Cats that urine mark are often agitated by outdoors cats and agonisitic interactions with other cats is correlated with feline urine marking.

The patient was also weaned off the Buspirone over the course of several weeks. Not only had it not been effective in changing behavior, but if the underlying causation was endocrine, anti-anxiety medication is not warranted.

Territorial behavior and aggression

The owner was also advised to confine the cat before company arrived with a special food treat or toy to avoid aggressive encounters.

Follow up

Contact with the owner one week after the surgery showed some urine spraying had occurred, but at a lower frequency than before surgery. When company is present the cat is confined or out on a leash and these two things have also curtailed urine spraying. The addition of Feliway was recommended since it can be effective in diminishing urine spraying by up to 75%.

Further follow up showed a decrease in urine marking and aggression over time. However when the cat was very agitated he might urine mark. Because it has been shown that even neutered animals will mark their territory the remaining urine marking was likely due to a learned component and frustration.

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REPORT OF LABORATORY EXAMINATION

Client: Owner:

Animal Health&Healing (30996)
2615 South Big Bend Boulevard
St. Louis, MO 63143 USA

Rec'd Date:	8/24/2006 12:22:00 PM	Animal:	MERLIN	MRN:	
Admitted By:	Snodgrass, Dr.	Species:	Feline	Breed:	Cat Mixed Breed
Ordered By:	N/A	Age:	11 years	Gender:	Male, Castrated
Encounter:	00219740	Tag/Reg ID:			
CR#:	GL	Other ID:			

Endocrinology

Endocrine Results

Collected Date/Time (If Provided)	08/23/2006 12:22:00		
Procedure		Ref Range	Units
Testosterone Baseline	3.5 H	[<=.5]	nmol/L
Total Thyroxine (TT4)	43	[10-55]	nmol/L
Total Triiodothyronine (TT3)	0.9	[0.6-1.4]	nmol/L
Free Thyroxine (FT4)	13	[10-25]	pmol/L
Free Triiodothyronine (FT3)	3.1	[1.5-6.0]	pmol/L
Endocrinology Interpretation	See Below		

08/23/2006 12:22:00 Endocrinology Interpretation
This concentration of testosterone is elevated for a netuered cat. Cryptorchidism remains a possibility, but I would have expected continuous male behavior from a young age. In the years that I have been here, we have seen 2 male cats with adrenocortical tumors that appear to have had testosterone as the main secretory product. In one of these cats, the male behavior and increased testosterone were alleviated with surgical removal of the tumor. In light of the testosterone result, the normal thyroid profile results do not support a differential diagnosis of hyperthyroidism.

Kent R. Refsal, DVM PhD
Professor, Endocrinology

L = Low Result; H = High Result; @ = Critical Result; ^ = Corrected Result; * = Interpretive Data; # = Result Footnote

DISEASE RESISTANCE IN SPOTTED HYENAS

WHAT'S IN THE BLOOD?

Contributing Authors:
Michele Miller, DVM, MPH
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The availability of field tests to provide rapid assessment of health status is critical to decision-making especially for translocation.

HYENAS often conjure up negative images due to their scavenging nature and have been associated with gluttony, cowardice and even black magic in some cultures. However, hyenas play an important ecological role especially as scavengers, by helping to recycle carrion inspiring the term "environmental health police". Once widespread throughout Africa, Arabia, Asia, and the Indian subcontinent, their conservation status is now considered near threatened due to habitat loss and human persecution. There are three species of hyena: spotted hyena (*Crocuta crocuta*), a powerful predator that hunts in packs and is found throughout sub-Saharan Africa; striped hyena (*Hyaena hyaena*), a mostly solitary scavenger with a range including Africa, the Middle East, and Western India; and brown hyena (*Parahyaena brunnea*), a smaller cousin of the spotted hyena that scavenges in packs or singly in the arid regions of southern Africa.

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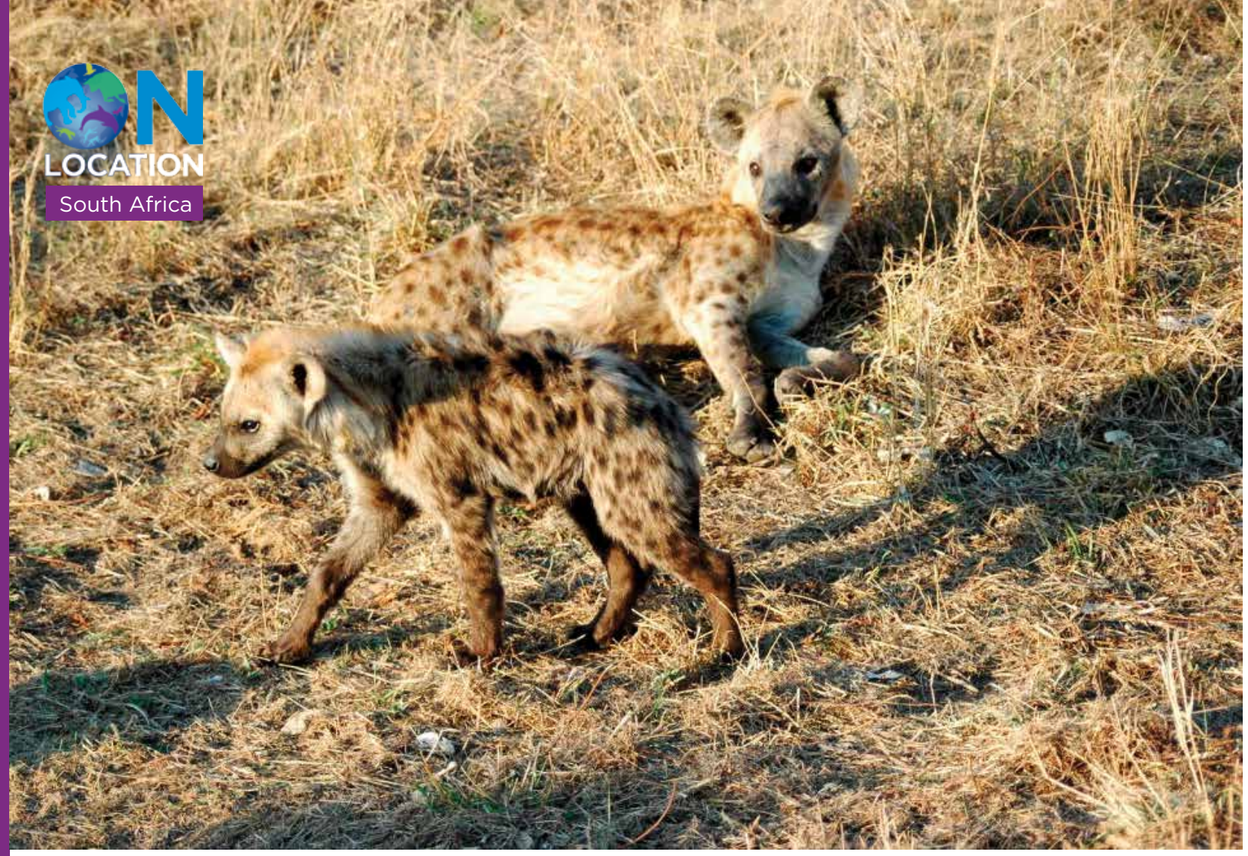

Two Spotted Hyenas

As a keystone predator in Africa, spotted hyenas have the greatest behavioral adaptation to habitat change, thus being indicators of ecosystem health. If a spotted hyena population declines or disappears, it suggests that the habitat has undergone severe degradation. Hyenas' other unique attributes warrant additional investigation such as their apparent resistance to diseases that can cause devastating effects on other carnivore populations such as rabies, canine distemper virus and bovine tuberculosis. Conservation programs to maintain and restore hyena populations in their current range are in progress. However, more information is required to assess health in animals that are being captured and potentially translocated. One of the primary principles of conservation is to minimize deleterious impacts such as introduction of disease. Therefore, the **Animal TB Research Group at Stellenbosch University in South Africa** along with the **Veterinary Wildlife Services staff in Kruger National Park**, have initiated a study to characterize the immune system and investigate its role in response to bovine tuberculosis in spotted hyenas. Our research focus has been to develop and evaluate a variety of techniques to determine health status, including biochemical panels using the **Abaxis comprehensive rotor.** One of the goals is to use this information to create a database that will facilitate disease detection in hyenas.

Very preliminary work has shown that spotted hyena have similar serum chemistry values compared to domestic dog parameters using serum on the Abaxis Comprehensive Rotor. Two healthy hyenas were being translocated from a facility without a history of bovine tuberculosis or other diseases. On physical examination, both were in excellent condition without any abnormalities. Blood samples from these individuals were used for a series of immunological tests as well as providing a baseline range for serum chemistry parameters. The availability of field tests to provide rapid assessment of health status is critical to decision-making especially for translocation. The chemistry results from the two hyena are shown below compared to the reference range provided by the Abaxis VS2 Chemistry Analyzer for the domestic dog.

Comprehensive Diagnostic Profile (CDP)

	Healthy hyenas	Dog normal range
Alb g/dl	2.3	2.5-4.0
ALP u/l	24-26	20-150
ALT u/l	105-109	10-118
Amy u/l	271-323	200-1200
Tbil mg/dl	0.4	0.1-0.6
BUN mg/dl	21-30	7-25
Ca mg/dl	10.1-10.3	8.6-11.8
Phos mg/dl	3.2-3.6	2.9-6.6
Cre mg/dl	2.0-2.3	0.3-1.4
Glu mg/dl	91-99	60-110
Na mmol/l	136-137	138-160
K mmol/l	4.8-5.1	3.7-5.8
TP g/dl	7.0-7.1	5.4-8.2
Glob g/dl	4.7-4.8	2.3-5.2

Spotted Hyenas scavenging a rhino carcass.

Hematology and biochemical values, along with clinical signs, and specific diagnostic tests being developed for TB in spotted hyenas will provide valuable information that can be used to assess health status in this species. Creating databases such as biochemical values adds to our knowledge of the biology of these unique animals.

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SIYABONA AFRICA | KRUGER NATIONAL PARK

Dr. Miller retrieving a blood sample from a Spotted Hyena.

OPERATION:
RESCUE BOROMOKO
AN ORPHANED BABY BULL

ON LOCATION
Serengeti

THE Mara Conservancy received a call from Brian Heath, the Chief Executive of the Mara Conservancy, about an abandoned baby elephant who had been observed for a couple of days on the plains of the Mara. Numerous elephants were in the area, moving through but this baby bull was seemingly an orphan because he never successfully integrated. Observation reported that this calf had not fed on any milk, and was very much peripheral to the herds, sometimes kilometers apart. The calf was left another day to see if anything changed and if his mother would return. There was a young female, too young to be his mother, who was clearly agitated and concerned about the little baby bull's fate and she was torn between remaining with the herds, and providing protection for the baby. It was thought that maybe she could be the sister of the little bull.

The David Sheldrick Wildlife Trust (DSWT) rescue team was called and alerted that the calf was getting weaker as he had not fed at all during observation, and if he were to remain unattended by the wild elephant herds he would fall prey to the predators. **It was now obvious he was an orphan**. The DSWT team flew to the Kichwa Tembo airstrip and was met and collected by Mara Conservancy Scouts and together with Brian Heath driven to the place the young calf still wandered the plains. On route to his location they passed plenty of hyenas and a pride of lions; which highlighted the fact that

continue ►

MARA CONSERVANCY
Protecting the Mara Triangle
www.maratriangle.org

The David Sheldrick Wildlife Trust

NAIROBI ORPHANS' NURSERY

Over the past three decades the Nairobi Nursery has successfully hand-raised over 150 orphaned elephant calves. These innocent baby elephants have been orphaned due to numerous reasons including poaching, human-wildlife conflict and natural disasters.

Contributing Authors:
David Sheldrick
Dr. Clay Wilson
Baerbel Koehler

VetScan VS2 Chemistry Analyzer and VetScan HM5 Hematology Analyzer in use on location.

ON LOCATION
Serengeti

he had survived the night and was extremely lucky. As they drove up close to where the baby calf was they could see the young female who had been most concerned about him, but even she was quite a distance away by this time. As the team leapt from the car to capture the baby the second landrover was strategically positioned to protect the men just in case the young female decided to charge. The little baby put up little resistance and was restrained in minutes. He was clearly very weak by this time. He was lain down on the stretcher, strapped and placed in the back of the landrover and immediately driven to the airstrip to the waiting aircraft. There at the airstrip we attempted to hydrate the baby calf. He was then loaded into the plane and flown to Nairobi, a journey of just under one hour.

He arrived in the DSWT Nursery late evening, the sweet little calf of approximately 15 months old settled down quickly and was relieved to have the company of the others and milk. The calf was hooked on his milk bottle almost immediately and with his pale amber eyes was instantly recognizable. After a complete blood chemistry and examination, it was only a couple of days before he was out with the others. We have called him Boromoko after the area where he was rescued. Boromoko loves to linger with the visiting foster parents in the evening when coming home to his stable and has to be tempted by his milk bottle in order to proceed to his stable. This is his unique little idiosyncrasy which of course endears him to everyone. The fate of his lost mother has not been confirmed, but there has been poaching reported in the Mara ecosystem in recent months.

Boromko's rescue and journey

Boromko's rescue and journey. *(From left to right.)*

Top:
Arriving at the Kichwa Tembo private airstrip.
Boromko being rescued by DSWT team and Mara Conservancy Scouts.

Middle:
Boromko being restrained with little resistance.
Transporting Boromko from the plains of the Mara to the airstrip.
Loading Boromko onto the aircraft before heading to Nairobi.

Bottom:
Boromko lingering with the visiting foster parents.

VETCOM would like to introduce the first spread of *PetTalks: Be the Voice For Your Pets*. Be a proud owner and shine some light on your pet for possible publication in any issue of Vetcom. We encourage you to get involved and share your PetTalk photos whether they may be funny, inspiring, or just too dang cute. We appreciate your continued readership and participation in Vetcom and look forward to seeing what your pets have to say!

It's as easy as 1, 2, 3. Here is what we're looking for:

1. Find a photo that captures your pet's unique look or odd behavior.
2. Imagine what your pet might be thinking and add a witty caption to the photo.
3. Email your PetTalk photo to **pettalks@abaxis.com** for possible publication.

Hercules

submitted by Kyle Holsworth
San Jose, CA.

Kelly

submitted by Matthew Souza
San Ramon, CA.

This is a cockatiel that our team treated for a broken foot. Recovered well.

Claude Morris | Morris Veterinary Hospital

PET TALKS

BE THE VOICE FOR YOUR PETS

Tai Shan at the National Zoo in DC, before he returned to China.

Suzanne Braden | Pandas International. photo by: Karen Wille

(Rescue cat)

submitted by Ken

Reggie

submitted by Simone

HERE ARE THE WINNERS!

We have been breeding Tennessee Walking Horse for over 40 years. All of our mares are sired by world champions and we always select world champion stallions for future breeding's. Several years ago we selected the stallion Walk Time Charlie to cross with Generator's Maid of Honor. The result was a beautiful colt with the markings of his sire and dam. He was a handful to say the least. Last year at the yearling production sale he was the second highest selling colt. Now under saddle he has sold again and attracted the attention of industry notables. We look forward to watching this colt continue to develop and cheer him on from the sidelines. His name, HRH Prince Charles!

Charlene B. Cook from Central Georgia Equine Service, Inc.

This is Chico. He is an adorable 7 year old Chihuahua that was rescued when a client abandoned him at our Veterinary office when he was only 4 month old. Even with this happening, he still has a positive outlook in life. He loves people, dogs, and especially cats. He also does not let his shortness get in the way. I think he is takes advantage of being so close to the ground and he is closer to getting anything that falls on the ground.

Susan Lopez from Pet Hospital

Curry, a smooth Brussels Griffon, is my constant companion. At work and at play, he is always willing. At work, he comforts sad clients and entertains children with his tricks. At home, he is friends with the horses, chickens and cats. He is an accomplished competitor in Obedience, Rally, Barn Hunt, K9 Nosework and Canine Freestyle. All this in a little 15 pound package.

Kate An Hunter

Young and eager, Hank is a cowdog in training. Working cattle is instinctual to him. He is tireless in helping gather, move and sort cattle. But a good cowdog must also learn to obey commands. Many times, the hardest job of all is following directions to stay put while the action continues all around. This shot caught him doing his very best to do just that. He had been in the midst of helping gather the cows and put them in the lot when he was called off, asked to load up in the truck, and stay. His expression shows how he longed to get back to work, but he followed orders. Good Dog.

Debra Jackson

Our Irish wolfhound puppy's first Christmas - Keely was 5 months old over Christmas - and definitely made the season brighter for our family! A new bear became a treasured toy.

Dr. Robyn Rodgers
Vermilion, Alberta

Frankie is one of the most interactive, engaging cats I've ever met. And he LOVES the camera! Picking his "best shot" was challenging because he is so photogenic, and practically smiles on cue when you take out the camera!

Julie from Carver Lake Veterinary Center

USDA & Canine Heartworm:

Understanding Low Worm Burden Testing

Contributing Authors:
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Rajesh Mehra, PhD | Director, Research & Development | Abaxis, Inc. | Union City, CA
Craig Tockman, DVM | Vice President, Sales and Marketing, NAAH | Abaxis, Inc. | Union City, CA

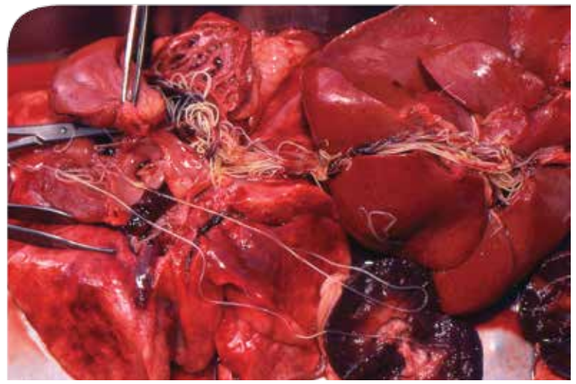
Background

Heartworm disease caused by infection with Dirofilaria immitis has been diagnosed in all 50 states[1, 2]. Testing for this parasite is routinely performed during regular checkups and annual visits as well as when clinical signs are present. Veterinarians count on the accuracy and precision of point-of-care antigen tests to provide accurate screening for the healthy patient as well as to assist with diagnosis of the patient suspected of infection when presenting with clinical signs.

Patients with heartworm infection may have as few as one worm or as many as several hundred. Some of the factors that determine this include environment, frequency of exposure, prevalence in the region and frequency of preventive use[1, 3]. Higher worm counts usually provide a higher antigen level for the tests to identify. Conversely, it is assumed that low worm counts may not provide sufficient antigen leading to a false negative test. However, worm burden is only one aspect that can lead to false negative results.

Severity of the disease is thought to be directly related to worm burden, length of infection and the response of the infected patient[4]. However, of equal, if not greater, importance is the activity level of the dog. A controlled study showed dogs that were infected by surgical transplantation with 50 heartworms and exercise-restricted took longer to develop clinical disease and developed less pulmonary vascular resistance than dogs with 14 surgically transplanted heartworms that were allowed moderate activity. This outcome was also evident in naturally infected dogs where, again, there was no correlation between the number of heartworms and pulmonary vascular resistance. Both of these scenarios indicate that the host-parasite interaction plays a significant role in the severity of disease[1]. Some patients, such as those not on a regular preventive and living outdoors in highly endemic areas, can have very high numbers of worms at the time of diagnosis. In cases of indoor patients with minimal exposure in areas of low prevalence, worm numbers are likely to be lower. So clearly a veterinarian may see high burden infections, low burden infections and infections at all levels in between.

Even with this knowledge, some practitioners desire to understand the sensitivity and specificity of heartworm tests at low worm burdens to compare available tests. To provide this information in a fair manner, Abaxis has performed a study to understand the test results at multiple levels of infection.



Materials and Methods

Serum from 25 known positive and 24 known negative patients, verified by antigen testing and necropsy, were purchased from the University of Florida College of Veterinary Medicine, Department of Infectious Diseases and Pathology. Each sample was identified with the number of female, gravid female, male, immature and dead worms in each patient. The samples were tested on 4 lateral flow devices according to manufacturer's instructions. The devices used were: IDEXX SNAP® Heartworm RT Testa (Serial # 09440-EG652), Heska Solo Step CHb (Serial # 204290), Synbiotics Witness® HWc (Lot No. 1101463) and Abaxis Vetscan® Canine Heartworm Rapid Test (Serial # 103144). Number of female worms ranged from 1 to 55 across the 25 patient samples.

Table 1

Results of Tests From Patients With 4 or Less Female Worms

4 Female Worms or Less					
	VetScan	SNAP	Solo Step	Witness	Total Positive
Positive	8	8	7	6	9
False Negative	1	1	2	3	
Sensitivity Values	89%	89%	78%	67%	

Summary data from all 25 known positive patients are shown in Table 2.

Table 2

Results From All Positive Patients

1-55 Female Worms or Less					
	VetScan	SNAP	Solo Step	Witness	Total Positive
Positive	23	23	21	20	25
Negative	24	24	24	24	24
False Negative	2	2	4	5	
Sensitivity Values	92%	92%	84%	80%	

Discussion

Antigen tests are highly specific and sensitive in diagnosing heartworm infection. It is important to remember that specificity and sensitivity values are valid only when the test is run according to the manufacturer's instructions, commonly found in the package insert for the product. They are available as point-of-care tests to be used in veterinary facilities. The USDA Center for Veterinary Biologics regulates all inhouse heartworm tests. Before any heartworm test can be sold, the performance of the test has to be demonstrated through a series of clinical studies showing acceptable performance in the following areas: accuracy, precision, ruggedness, sensitivity and specificity. The submission data is the basis of the package insert for these tests and tests cannot be sold without meeting the USDA criteria. The Abaxis Vetscan Canine Heartworm Rapid Test meets or exceeds all these USDA standards[5]. Specificity is often perceived to mean accuracy, however, specificity is by definition the measurement of the frequency with which a result will be negative (or normal) in patients without the disease. To clarify, a test with 98% specificity would mean that if 100 negative or normal patients are tested, then the test would in fact show negative or normal in 98 of them, while 2 of the patients would show a false positive. Therefore a test with high specificity rules out the disease with a high level of certainty. Specificity is calculated as True Negatives/ (True Negatives + False Positives).

Immunoassays such as those for canine heartworm antigen are very specific to variations among individuals. Not all individuals have identical immune responses. Likewise, all infections are not identical and may be modulated by the health of the host or a variety of other factors. In addition, whether based on polyclonal or monoclonal technologies, the antibodies for different antigen tests may be specific to different antigens or even different epitopes of a common antigen, and normally differ among different manufacturers[6]. These are only some of the reasons that any test is capable of producing a false positive or false negative heartworm test regardless of the worm burden of the patient. To be led to believe anything else is dishonest at best.

Some companies aggressively market the accuracy of tests at low worm burdens, considered by some to mean 4 female worms or less, to suggest that their tests are more accurate than others, in spite of the fact that many heartworm infections involve much higher worm burdens along with clinical signs.
Several papers or studies have been presented to the veterinary community describing these results, with significant skewing of those results towards the publishing company's product[6].

The practitioner should also consider the positive and negative predictive value of each test they perform. Positive predictive value is the percentage of patients with a positive test that actually have the disease in question (True Positives) and includes either prevalence data or in the clinical setting, the likelihood of the disease based on history, physical examination and clinical signs. Negative predictive value is the percentage of patients who test negative to a disease and are in fact negative (True Negatives), but it also accounts for prevalence data, clinical illness or the lack thereof. Prevalence is the percentage of true positive animals within the tested population. When interpreting the diagnostic result, it is imperative to include either prevalence data for a screening protocol, or history and clinical signs in an ill patient[2, 7].

USDA & CANINE HEARTWORM: UNDERSTANDING LOW WORM BURDEN TESTING
HEARTWORM AWARENESS MONTH

The results of this study show all tests to have excellent specificity. It also clearly shows that all 4 of the tests, while good to excellent at identifying infection at all worm burdens, will generate an occasional false negative result. Practitioners must understand that this is a possibility with all tests and any company claiming to be perfect every time with every test is not being honest with the customer. A veterinarian with a patient exhibiting clinical signs of disease in the face of a negative test should confirm his/her clinical suspicions with a test of a different methodology as recommended by the American Heartworm Society[1]. Conversely, a patient which is not exhibiting clinical signs of disease in the face of a positive test should also be retested for confirmation with a different methodology.

The study also showed that the Abaxis Vetscan Canine Heartworm Rapid Test provided excellent results in comparison to all competitive brands across all worm burdens.

Table 3

Complete Experimental Data

ID	Worm Female	Gravid	Worm Male	Worm Immature	Worm Dead	Worm Total	VetScan	SNAP	Solo Step	Witness
2011308	45	Y	25	2	2	70	+	+	+	+
2011315	3	N	3	0	1	6	+	+	+	-
2011332	2	Y	1	0	1	3	+	+	+	+
2011340	4	Y	5	0	0	9	+	+	+	+
2011341	16	Y	12	0	0	28	+	+	+	+
2011342	3	Y	3	0	0	6	-	-	-	+
2011344	8	Y	10	0	0	18	-	-	-	-
2011347	46	Y	30	2	1	76	+	+	+	+
2011354	11	Y	4	0	2	15	+	+	+	+
2011355	33	Y	16	0	2	49	+	+	+	+
2011356	17	Y	26	0	1	43	+	+	+	+
2011357	20	Y	31	0	3	51	+	+	+	+
2011358	15	Y	12	0	1	27	+	+	+	+
2011365	3	Y	2	2	0	5	+	+	+	+
2011366	19	Y	9	0	1	28	+	+	+	+
2011371	15	Y	16	0	1	31	+	+	+	+
2011372	3	Y	1	0	0	4	+	+	+	+
2011373	55	Y	56	0	2	111	+	+	+	+
2011376	3	Y	4	0	2	7	+	+	+	-
2011377	10	N	0	0	1	10	+	+	+	+
2011384	10	Y	9	0	1	19	+	+	-	-
2011388	49	Y	40	2	5	89	+	+	+	+
2011389	2	Y	5	0	1	7	+	+	+	+
2011390	3	Y	7	0	1	10	+	+	-	-
2011391	25	Y	21	0	2	46	+	+	+	+

[a]SNAP®, IDEXX Laboratories, Portland, ME
[b]Solo Step CH, Heska Corp, Fort Collins, CO
[c]Witness® HW, Synbiotics, Kansas City, MO
[d]Vetscan® Canine Heartworm Rapid Test, Abaxis, Inc., Union City, CA

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