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FEATURE ARTICLE

The Importance of Routine Fecal Exams

Protecting pets and their owners from parasitic infections

Dwight D. Bowman
MS, PhD
Professor of Parasitology, Cornell University College of Veterinary Medicine

Araceli Lucio-Forster
BS, PhD
Teaching Support Specialist, Cornell University College of Veterinary Medicine

Why fecal exams are still required for pets

Human intestinal parasites in the U.S. have been virtually eradicated by improvements in sewage management, availability of clean drinking water, control of zoonotic parasites in food animals and the routine anthelmintic treatment of the human population. Dogs and cats have not yet been so fortunate. Byron L. Blagburn, MS, PhD, of Auburn University College of Veterinary Medicine, has conducted surveys showing that the level of infection among shelter dogs is similar to what it was 10 years ago, when the national average was more than 10% infected with hookworms, roundworms or whipworms.[1] An ongoing national survey has also found more than 25% of cats have roundworms and more than 10% have hookworms.[2]

Even when pets are well taken care of and live in nice neighborhoods, there may be other animals (e.g., strays, foxes, coyotes) infected with parasites defecating in their yards and parks. Normal behaviors, such as eating directly off the ground, drinking out of puddles, grooming, and playing fetch and pouncing and biting items on the ground, predispose dogs and cats to parasitic infections all year long. Dogs and cats also acquire infections from infected prey (e.g., rabbits or mice) or fleas that serve as intermediate or paratenic hosts for different parasites.

Year-round internal parasite prevention products do not guarantee that a pet will not have intestinal parasites. Recurring developmental stages of parasites, such as hookworms, periodically repopulate the intestine, grow to adulthood and shed eggs in feces. Eggs and cysts produced by parasites may not be susceptible to prevention or treatment by the products being administered. Lastly, owner compliance in administering preventatives may be lacking.

Protecting your clients from zoonotic disease

In addition to having health implications for the pet itself, some of the parasite stages shed in dog and cat feces are zoonotic, so removing them to prevent environmental contamination, thereby protecting clients and their families, is important. Cutaneous larva migrans by hookworms cause nasty skin lesions. Larval stages of ascarids can migrate through the liver, lungs and eyes causing organ damage and blindness respectively. With an estimated 3–6 million people infected each year, ascarid infection is one of the leading causes of unilateral blindness.

How often a fecal examination should be performed

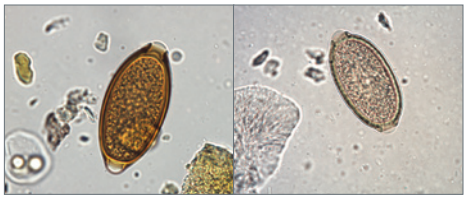
A pet's history and lifestyle gives some indication as to whether they are more or less likely to have certain parasites. Important factors include age of pet, recent adoption from a shelter, frequenting of dog parks, administration of year-round preventives, hunting, presence of fleas, access to rodents or cockroaches and presence of other medical problems. Even predominantly indoor pets are likely to be exposed to parasites.

The Companion Animal Parasite Council (CAPC) recommends that appropriate fecal examinations be performed on puppies and kittens at least 2–4 times during the first year of life and 1–2 times per year in adults (see insert for CAPC guidelines).

Factors influencing the reliability of a fecal examination

Some of the more important factors influencing the results of fecal examinations are the size, handling and age of the sample, choice of procedure, specific procedural methods, training of personnel conducting the procedure and the utilization of ancillary fecal diagnostics.

Samples need to be of adequate size and handled appropriately. For companion animals, 3–5 grams of material is ideal, while 1 gram is the minimal amount. Fresh fecal samples are required because the stage of the parasite (e.g., cyst, single-cell stage, morula stage, larval stage) in fresh feces is characteristic: a thick-shelled egg containing a single cell is characteristic of ascarid nematodes (roundworms), while thin-shelled morula-stage eggs are characteristic of hookworms. However, as



***Trichuris vulpis* (left) and *Eucoleus* (*Capillaria*) *aerophilus* (right) eggs are lemon shaped with bipolar plugs and are similar in appearance.**

soon as eggs and cysts are passed in feces, they begin to develop; for example, single-celled eggs become morulated, morulated eggs become larvated and larvated eggs can hatch, and these more mature parasite stages can be difficult to identify.

The procedure(s) chosen for fecal examination will depend to some extent on which parasites are suspected. Routine fecal examinations are usually done with a floatation procedure. All floatation procedures rely on the same basic principle: a salt (usually zinc sulfate or sodium nitrate) or sugar solution is used to effectively float eggs to the surface, which occurs because eggs are lighter than the specific gravity of these solutions. However, not all flotation methods are equal, with some procedures having better recoveries than others.[3] For example, those procedures that rely on gravity to do the separation between the parasitic stages and the debris ("standing" flotations) tend to have poorer recoveries than those using centrifugation. Thus, centrifugal fecal flotations are the gold standard when it comes to routine fecal examination.[3] A wet mount (a small amount of feces, may be diluted with saline, under a coverslip) to detect protozoan trophozoites should be considered for liquid stools. The Baermann method is useful for the detection of live larvae present in feces (e.g., feline lungworm). Antigen-detecting ELISAs are routinely used as patient-side or plate tests, most commonly for detection of *Giardia* and *Cryptosporidium*. Molecular methods, such as real-time PCR assays, are being used more and more for detecting fecal pathogens (see the following case study).

The importance of well-trained personnel to conduct and read the tests cannot be overstated. There are plenty of fecal artifacts that resemble parasites and can confuse the untrained.[4] The task of performing a fecal examination should be undertaken with care and pride. The prepared slides must be read in their entirety, in a methodical way, and at a magnification of at least 100× (otherwise, protozoan parasites and other small eggs may be missed or misidentified).

Summary

We can only hope that someday the task of fecal examination will become exceedingly boring because we are no longer finding anything. The goal is a negative sample so that, as occurs with people, tests will only be ordered when there is clinical suspicion of infection or a history supporting a need for a fecal examination. However, for now, fecal examinations continue to be an important component of the routine examination in our patients, and it is important to perform these tests in a manner that maximizes detection of parasitic infections. |DX|

REFERENCES

1. Blagburn BL, Lindsay DS, Vaughan JL, Rippey NS, Wright JC, Lynn RC, Kelch WJ, Ritchie C, Hepler DI. Prevalence of canine parasites based on fecal flotation. *Compend Contin Educ Pract Vet.* 1996;18(5):483–509.
2. Stockdale HD, Little SE, Blagburn BL. Canine and feline intestinal parasite prevalence revisited. Paper presented at: 53rd Annual Meeting of the AAVP; July 19–22, 2008; New Orleans, LA.
3. Dryden MW, Payne PA, Ridley R, Smith V. Comparison of common fecal flotation techniques for the recovery of parasite eggs and oocysts. *Vet Ther.* 2005;6(1):15–28.
4. Lucio-Forster A, Bowman DD. Differentiation of parasites and pseudoparasites. *NAVC Clinician's Brief.* 2009;7(4):54–58.

Everyday Fecal Diagnostics

Routine parasitic screening and identifying infectious causes of diarrhea

Jane Robertson, DVM, DACVIM

The preceding article, *The Importance of Routine Fecal Exams,* by Drs. Lucio-Forster and Bowman, highlights the importance of properly performing routine ova and parasite fecal examinations in companion animals. Fecal floatation by centrifugation performed by a well-trained technician is the gold standard for screening for common intestinal parasites in dogs and cats. On a wellness examination, the aim is to identify occult parasitic infections that may be causing unseen disease or have the potential to cause disease in the pet and may pose a zoonotic threat to the pet owner.

Fecal testing should also be performed early in the diagnostic workup of animals with diarrhea. Initial screening with fecal floatation by centrifugation and a *Giardia* ELISA is recommended. If infection is identified, the pet should be treated appropriately and monitored for resolution of clinical signs. If results are negative, other intestinal infections should be excluded prior to performing a more exhaustive and expensive diagnostic workup.

Recently, it has become easier and more economical to screen for multiple fecal pathogens. Available through IDEXX Reference Laboratories are the diarrhea RealPCR™ panels, which include real-time PCR assays for *Clostridium perfringens* enterotoxin A gene, *Cryptosporidium* spp., *Giardia* spp. and *Salmonella* spp., with the addition of canine distemper virus, canine enteric coronavirus and canine parvovirus 2 (CPV-2) in the canine panel and feline coronavirus (FCoV), feline panleukopenia virus, *Tritrichomonas foetus* and *Toxoplasma gondii* in the feline panel.

The following case study, by Dr. Aronson Webb, of a young golden retriever with chronic diarrhea, highlights the practical use of the diarrhea RealPCR panel for identifying an infectious cause of diarrhea and subsequently resulted in successful treatment.

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Chronic Diarrhea

The value of diarrhea RealPCR™ panels

Judy Aronson Webb
DVM

Willie

Patient: Willie, 5-month-old, neutered male golden retriever

Presenting reason: Chronically soft stools with episodes of runny diarrhea

History: Active, happy puppy adopted at 2 months of age. Soft to runny stools twice daily with no blood or mucous. Appetite good and growing normally. History of ongoing dietary indiscretion (i.e., ingestion of dirt, toys and sea water). Coccidia diagnosed on fecal floatation at 2 months of age; no response to treatment with Albon® and deworming with pyrantel pamoate. Fecal ova and parasite examinations and *Giardia* ELISAs at 3 and 4 months of age were negative. Flea, tick, and heartworm prophylaxes have been administered monthly.

Initial examination

Physical examination

Willie was bright, alert, responsive and hydrated. His body condition score (BCS) was 5/9. Abdominal palpation revealed no abnormalities. His skin and hair coat looked healthy. The remainder of the physical examination was within normal limits.

Assessment

Since Willie was otherwise healthy and growing normally his diarrhea was considered most likely large bowel in nature; however, there was no increase in frequency or urgency and no blood or mucous to support this. In spite of repeated negative fecal examinations, but given his age, an infectious cause of his diarrhea was still considered most likely.

Diagnostic Plan

A complete blood count (CBC), clinical chemistry profile, fecal ova and parasite examination by a centrifugal floatation technique and a *Giardia* ELISA were performed.

CLINICAL CHEMISTRY PROFILE	VALUE	UNITS		REF INTERVAL
Glucose	107	mg/dL		(60 – 125)
BUN	11	mg/dL		(7 – 27)
Creatinine	0.7	mg/dL		(0.4 – 1.8)
B/C Ratio	15.7			
Phosphorus	9.0	mg/dL	High	(2.1 – 6.3)
Calcium	10.9	mg/dL		(8.2 – 12.4)
Sodium	149	mmol/L		(141 – 156)
Potassium	5.2	mmol/L		(4.0 – 5.6)
Na/K Ratio	29			(27 – 40)
Chloride	108	mmol/L		(105 – 115)
tCO_2 (Bicarbonate)	19	mmol/L		(17 – 24)
Anion Gap	27	mmol/L	High	(12.0 – 24.0)
Total Protein	5.1	g/dL		(5.1 – 7.8)
Albumin	2.0	g/dL	Low	(2.5 – 4.0)
Globulin	3.1	g/dL		(2.1 – 4.5)
A/G Ratio	0.6			(0.6 – 1.1)
ALT	26	U/L		(5 – 107)
AST	34	U/L		(5 – 55)
ALKP	225	U/L	High	(10 – 150)
GGT	5	U/L		(0 – 14)
Total Bilirubin	0.1	mg/dL		(0.1 – 0.4)
Direct Bilirubin	0.0	mg/dL		(0.0 – 0.1)
Indirect Bilirubin	0.1	mg/dL		(0.0 – 0.3)
Cholesterol	210	mg/dL		(112 – 328)
Amylase	626	U/L		(350 – 1050)
Lipase	263	U/L		(100 – 750)
CK	263	U/L	High	(10 – 100)
Hemolysis Index[1]	9			
Lipemia Index[2]	9			

Comments:
1. Index of <300 exhibits no clinically significant effect on chemistry values.
2. Index of <500 exhibits no significant effect on chemistry values.

HEMATOLOGY	VALUE	UNITS		REF INTERVAL
RBC	5.28	M/μL	Low	(5.50 – 8.50)
HCT	38.1	%		(37.0 – 55.0)
HGB	12.6	g/dL		(12.0 – 18.0)
MCV	72.0	fL		(60.0 – 77.0)
MCH	23.8	g/dL		(19.5 – 26.0)
MCHC	33.0	g/dL		(32.0 – 36.0)
WBC	11.2	K/μL		(5.7 – 16.3)
Neutrophil	51.52	K/μL		(3.00 – 11.50)
Lymphocyte	4.70	K/μL		(1.00 – 4.80)
Monocyte	0.67	K/μL		(0.15 – 1.35)
Eosinophil	0.67	K/μL		(0.10 – 1.25)
Basophil	0.0	K/μL		(0.0 – 0.1)
PLT (Automated)	235	K/μL		(164 – 510)

Remarks: Slide reviewed microscopically

ADDITIONAL DIAGNOSTICS	RESULT
Fecal Ova and Parasites	No ova and parasites seen
Giardia ELISA	Negative

Laboratory findings

The mild elevation of serum phosphorus and alkaline phosphatase (ALKP) were considered normal for a 5-month-old puppy. His hypoalbuminemia was likely secondary to gastrointestinal loss, which did suggest a small bowel component

to his diarrhea. Bile acids to rule out a liver shunt and a urinalysis to rule out proteinuria were advised. The increased anion gap, especially in light of a decreasing albumin that will result in a decrease in the anion gap, suggests the presence of a titrational metabolic acidosis (increase in unmeasured anion). However, there are no obvious clinical signs, physical findings or other laboratory abnormalities that support this interpretation. Repeat electrolyte and acid-base assessment is warranted following correction of the primary clinical problem.

Assessment

A mild protein-losing enteropathy of unknown etiology was the tentative diagnosis at this time. A parasitic cause was considered unlikely based upon repeated negative fecal examinations and treatment with a monthly internal parasite preventative.

Therapeutic plan:

Trial therapy with metronidazole, probiotics and bland diet.

Representation 2 years later

History

Continues to have soft to runny stools. Responds to treatment with metronidazole, but diarrhea relapses when off therapy. No response to various dietary trials. Repeat fecal ova and parasite examination and *Giardia* ELISA 3 months earlier were negative. Otherwise, Willie was a happy, healthy full-grown dog.

Physical examination

Physical examination was within normal limits. BCS was 5/9.

Assessment

Differential diagnoses for chronic diarrhea include infectious causes (e.g., parasitic, fungal, bacterial, viral, protozoal), dietary sensitivity and inflammatory bowel disease.

Diagnostic plan

A repeat CBC and chemistry panel were recommended but not performed at this time. A repeat ova and parasite examination and *Giardia* ELISA were performed. In addition, a diarrhea RealPCR™ panel that had recently become available was performed to look for additional infectious causes of diarrhea. During this visit, it was also discussed that if these fecal tests did not identify an infectious cause to explain Willie's chronic diarrhea, then a more extensive workup would be recommended.

ADDITIONAL DIAGNOSTICS	RESULT
Fecal Ova and Parasites	No ova and parasites seen
Giardia ELISA	Negative

CANINE DIARRHEA RealPCR™ PANEL	RESULT
Canine distemper virus	Negative
Canine enteric coronavirus	Negative
Canine parvovirus 2 (CPV-2)	Negative
Clostridium perfringens enterotoxin A gene	Negative
Cryptosporidium spp.	**Positive**
Giardia spp.	Negative
Salmonella spp.	Negative

A positive *Cryptosporidium* spp. result indicates that *Cryptosporidium* spp. DNA was detected in the fecal sample. In animals with clinical signs, this supports infection. Clinical signs include acute, chronic or intermittent small- and/or large-bowel diarrhea. Positive animals can be asymptomatic at time of testing but are still a source of environmental contamination. Zoonotic potential may exist.

A negative *Cryptosporidium* spp. result indicates that *Cryptosporidium* spp. DNA was not detected in the fecal sample and suggests that *Cryptosporidium* spp. is not the cause of diarrhea in this animal. However, negative results can result from previous antiprotozoal therapy, levels of organisms being too low for detection or 100% of clinically important isolates not being detected.

Laboratory findings

The diarrhea RealPCR panel was positive for *Cryptosporidium* spp. *Cryptosporidium* infection can cause acute/chronic/intermittent small- and/or large-bowel diarrhea, and a positive PCR result for this organism was considered significant.

Therapeutic plan

Although, there is no treatment that is considered 100% effective for *Cryptosporidium* infection, many dogs respond favorably to treatment with azithromycin. Willie was treated with azithromycin at 10 mg/kg twice daily for 10 days.

Clinical case outcome

After two days on therapy Willie's stools became normal. Ten months later, he continues to have normal stools.

This case highlights the diagnostic utility of the diarrhea RealPCR panel to help identify infectious causes of diarrhea that are difficult or expensive to detect by conventional methods. In a young dog with a chronic history of diarrhea, an infectious cause is considered most likely and the diarrhea RealPCR panel should be considered after routine screen with a fecal ova and parasite examination performed by a centrifugal floatation method and a *Giardia* ELISA. In this case, identifying and treating Willie's *Cryptosporidium* infection prevented further unnecessary diagnostics, such as endoscopy, that were being considered in an attempt to diagnose the cause of his diarrhea.