Studies show that Fecal Dx antigen tests allow for earlier detection of more intestinal parasites



Antigen detection is commonly used today to diagnose heartworm and *Giardia* infections, and now it is available for additional parasites. IDEXX Reference Laboratories, as a leader in pet healthcare innovation, has developed immunoassays for the detection of hookworm, roundworm, and whipworm antigens in feces of dogs and cats. These antigens are secreted from the adult worm and are not present in their eggs, which allows for detection of prepatent stages as well as the ability to overcome the challenges of intermittent egg laying. Earlier detection during the prepatent period will also reduce the frequency of environmental contamination with potentially infectious eggs.

Two recent papers describing the performance of the Fecal Dx[™] antigen tests, enzyme-linked immunosorbent assays (ELISAs) developed by IDEXX for coproantigen detection of *Trichuris vulpis*, *Ancylostoma caninum* and *Toxocara canis* in dogs and *Toxocara cati* in cats, are summarized below.

- Enzyme-linked immunosorbent assay for coproantigen detection of *Trichuris vulpis* in dogs¹
- Enzyme-linked immunosorbent assays for coproantigen detection of *Ancylostoma caninum* and *Toxocara canis* in dogs and *Toxocara cati* in cats²

Purpose

The purpose of these studies was to confirm the utility of coproantigen detection for diagnosing *T. canis*, *A. caninum*, and *T. vulpis* infections in dogs and *T. cati* infections in cats.

Study designs

Fecal samples from naturally infected dogs and cats and from experimentally infected dogs were tested with both fecal flotation by centrifugation and individual ELISAs (Fecal Dx antigen tests) specific for coproantigen for roundworms, hookworms, and whipworms.

To evaluate samples from naturally infected animals, data from fecal flotation and the Fecal Dx antigen tests was collected from approximately 1,000 field canine and feline fecal samples submitted to IDEXX Reference Laboratories. An additional immunoprecipitation assay was developed to confirm the specificity of the Fecal Dx antigen test in those fecal samples that were egg negative and antigen positive.

In addition, a total of 15 dogs, 5 dogs in each group, were experimentally infected with either *T. canis*, *A. caninum*, or *T. vulpis* to confirm the detection of the prepatent period by each assay.

Results

In the 1,156 field fecal samples for the roundworm and hookworm study and the 1,000 field fecal samples for the whipworm study, egg-positive roundworm, hookworm, and whipworm results were noted in 23, 13, and 27 samples, respectively. In contrast, 26, 19, and 35 samples were antigen positive for roundworm, hookworm, and whipworm. The *T. canis* ELISA detected *T. cati* coproantigen in feline samples. Fecal antigens detected more infections than did fecal flotation.

	Roundworm	Hookworm	Whipworm
Fecal flotation positive	23	13	27
Fecal Dx antigen test positive	26	19	35

Table 1. The number of nematode-positive fecal results from fecal samples submitted to IDEXX Reference Laboratories (n = 1,000 for roundworm and hookworm, n = 1,156 for whipworm)

Discrepant Fecal Dx antigen test and flotation results were obtained for 13 roundworm, 16 hookworm, and 12 whipworm positive samples that were negative by the alternate method. After reexamination of the discrepant samples that were egg positive but antigen negative, several instances of probable misidentification or coprophagy were noted. Conversely, the specificity of the Fecal Dx antigen test in the field samples that were antigen positive but egg negative was confirmed with the immunoprecipitation assays for each of the three nematodes.

Experimental infection studies confirmed detection of the prepatent stages of all three nematode parasites. Postinfection coproantigen was detected as early as day 31 for *T. canis*, day 9 for *A. caninum*, and day 23 for *T. vulpis*. Eggs were not detected by flotation until day 38, 23, and 69, respectively. Fecal antigens diagnosed infections earlier than egg detection (see table 2).

	Nematode detection (days postinfection)			
	Roundworm	Hookworm	Whipworm	
Fecal flotation	38	23	69	
Fecal Dx antigen test	31	9	23	

Table 2. The day of appearance of positive fecal results in dogs following experimental nematode infection (n = 15).

The experimental infection study also confirmed that the coproantigen was specific for each individual nematode. In addition, specificity of the assay was confirmed by lack of cross-reaction with closely related nematodes that were tested, e.g., capillarids for whipworm.

Conclusion

Fecal Dx[™] antigen tests detect more parasitic infections and detect infections earlier than fecal flotation. Flotation, while familiar and accessible, has challenges and may not accurately identify all infections. In many field samples, there was good agreement between fecal flotation and antigen test results. However, as these methods are testing for different attributes, presence of eggs versus presence of coproantigen from an adult worm, differing results are to be expected. There was a subset of samples found to be egg positive and antigen negative and another subset found to be egg negative and antigen positive. As seen with egg-negative and antigen-positive samples, an advantage of coproantigen testing is the ability to detect prepatent stages before egg shedding. This was confirmed in the experimental infection studies. As the authors of these publications have noted, it is known that cats and dogs receiving monthly anthelmintics effective against roundworms and hookworms may test positive for adult worms within the month, often through coproantigen detection and rarely by egg identification on flotation. This is attributed to the relatively short prepatent periods of these nematodes and a variety of infection scenarios, such as maturation of arrested larvae from tissues, ingestion of Toxocara eggs or Ancylostoma larvae, ingestion of paratenic hosts like mice, or skin penetration of hookworm larvae. Because of the continued risk of infection in these pets, it is particularly important that they continue to receive a broad-spectrum monthly anthelmintic year-round to prevent the continued development of worms and subsequent shedding of eggs. Finally, coprophagy may be a common habit in many dogs, and reexamination of egg-positive and antigen-negative canine samples also confirmed the presence of spurious eggs, likely from the ingestion of infected feces.

The additional diagnostic capabilities of the Fecal Dx antigen tests for roundworms, hookworms, and whipworms fill the gaps found with fecal flotation and enable practitioners to diagnose more intestinal parasites earlier in the infection cycle, allowing for timely treatment and reducing environmental contamination with potentially infectious, zoonotic eggs.

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References

- Elsemore DA, Geng J, Flynn L, Cruthers L, Lucio-Forster A, Bowman DD. Enzyme-linked immunosorbent assay for coproantigen detection of *Trichuris vulpis* in dogs. *J Vet Diagn Invest*. 2014;26(3):404–411.
- 2. Elsemore DA, Geng J, Cote J, Hanna R, Lucio-Forster A, Bowman D. Enzyme-linked immunosorbent assays for coproantigen detection of *Ancylostoma caninum* and *Toxocara canis* in dogs and *Toxocara cati* in cats. J Vet Diagn Invest. In press.

