Probiotics or pathogens? Unraveling the role of intestinal bacteria in kitten diarrhea

Jody L. Gookin, DVM, Ph.D., DACVIM
Raleigh, NC

INTRODUCTION

Approximately ~82 million owned[1] and 70 million feral[2] cats currently reside in the United States. Population projections estimate that these cats give birth to roughly 180 million kittens per year[3]. Humane efforts focusing on high-volume adoption are placing record numbers of healthy kittens into homes. However, the death or euthanasia of ailing kittens remains a national tragedy[4-11]. Each year inestimable numbers of kittens are abandoned, orphaned, or relinquished shortly after birth. While the exact statistics are unknown, ~15% of these kittens will die or be euthanized because of illness before they reach 8-weeks of age[12,13]. Kittens (and puppies) are the easiest-to-place pets in animal shelters.[13] Accordingly, improving the health and welfare of these kittens will place more shelter kittens into homes[12,13], reduce the number of kittens euthanized in animal shelters, and advance the humane ethic of the veterinary profession. We know distressingly little about why so many foster kittens die. While infectious causes are suspected to be most likely,[14-16] studies reporting the prevalence of infectious agents in this population are limited[14]. In theory, insufficient intake of colostrum by fostered kittens will lead to failure of passive transfer (FPT)[17,18] and increased susceptibility to infectious disease. However efforts to document the presence or clinical significance of naturally-occurring FPT in kittens have not been reported. An obvious cause of death is unknown in as many as 53% of kittens[7], however many are reported to have clinical signs of diarrhea[7,12,19] or post-mortem evidence of enteritis.[14] For these reasons, research aimed at identifying previously unrecognized infectious agents of the GI tract in foster kittens and those that significantly contribute to mortality is likely to be a productive focus of investigation.

INFECTIOUS CAUSES OF GASTROINTESTINAL DISEASE IN THE KITTEN

The following are common infectious causes of gastrointestinal disease in kittens and the multitude of diagnostic tests available for use in their identification. In many instances the relative predictive value of these tests remain unknown.

Table 1. Common infectious causes of diarrhea in young kittens

Worms	**Recommended Diagnostic test(s)**	**Diagnostic finding(s)**	**Treatment**
Hookworms, Whipworms, Roundworms etc	Centrifugation- Flotation	Ova	Fenbendazole
Ollulanus, Physaloptera	Microscopic examination of vomit	Worms	
	Fecal sedimentation	Ova	
Viruses	**Diagnostic test(s)**	**Diagnostic finding(s)**	**Treatment**
Feline *Panleukopenia*	CPV ELISA	Antigen positive	Supportive
	Fecal PCR	DNA positive	
Feline enteric *Coronavirus*	Fecal RT-PCR	mRNA positive	
	Serology	Antibody positive	
Feline *Astrovirus*	Fecal TEM	Virus particles	
	Fecal PCR	Fecal DNA	

Table 1. Common infectious causes of diarrhea in young kittens			
Protozoa	**Diagnostic test(s)**	**Diagnostic finding(s)**	**Treatment**
Cystoisospora	Centrifugation-flotation	Ova	Ponazuril
Giardia	Centrifugation-flotation	Oocysts	Fenbendazole, Metronidazole
	ELISA	Antigen positive	
	Fecal PCR	DNA positive	
Cryptosporidium	Centrifugation-flotation	Oocysts	None proven (Nitazoxanide?)
	Cytology	Acid-fast positive	
	IFA (Merifluor)	Antigen positive	
	ELISA		
	Fecal PCR	DNA positive	
Tritrichomonas	Fecal wet mount	Trophozoites	Ronidazole
	Fecal culture InPouch^{TF}	Trophozoites	
	Fecal PCR	DNA positive	
Toxoplasma gondii	Centrifugation-flotation	Ova	Clindamycin
	Fecal PCR	DNA positive	
Bacteria	**Diagnostic test(s)**	**Diagnostic finding(s)**	**Treatment**
Salmonella	Fecal culture x 3	*Salmonella*	Supportive ± antibiotics
	PCR	DNA positive	
Campylobacter	Fecal culture	*Campylobacter spp.*	Erythro/Azithromycin
	Species-specific multiplex PCR	Pathogenic *Campylobacter*	
Clostridium difficile	Culture or PCR	*C. difficile*	Metronidazole
	ELISA (TcdA, TcdB)	"Toxigenic" *C. difficile*	
Clostridium perfringens	Fecal culture	*C. perfringens*	Amoxicillin, metronidazole, or tylosin
	Fecal PCR for cpe gene	"Toxigenic" *C. perfringens*	
	Fecal ELISA for CPE antigen		
Diarrheagenic *Escherichia coli*	Virulence gene PCR on culture isolate of feces		Antibiotics based on sensitivity testing
	eae, stx1, stx2	Enterohemorrhagic *E.coli*	
	eae, bfp	Enteropathogenic *E.coli*	
	STa, STb, LT	Enterotoxigenic *E.coli*	
	CNF1, CNF2	Necrotoxigenic *E.coli*	

EXAMPLE OF A DIAGNOSTIC PLAN

There are many diagnostic tests that are useful for identifying infectious causes of gastrointestinal disease and for many of these agents there are multiple tests. The following represents the author's general diagnostic approach to ruling out infectious causes of gastrointestinal disease in the kitten.

Diagnostic Tests	**Core Database**	**Prioritize if systemically ill or has acute, hemorrhagic, or febrile diarrhea**
Complete physical examination	+	
Fecal wet mount(s)	+	
Fecal centrifugation flotation	+	
Examination of vomitus for parasites	+	
Giardia antigen test		
Tritrichomonas foetus test		
Complete blood cell count		+
Fecal cytology		+
Parvovirus fecal ELISA or *Panleukopenia* PCR		+
Fecal culture or PCR for *Salmonella, Campylobacter*, pathogenic *E.coli*		+
Cryptosporidium IFA, ELISA or PCR		+
Clostridium culture/PCR/ELISA		+

PREVALENCE OF BACTERIAL ENTEROPATHOGENS AND ASSOCIATIONS WITH DIARRHEA

Enteropathogens prevalence only

The Veterinary Journal 201 (2014) 189–195

Contents lists available at ScienceDirect

The Veterinary Journal

journal homepage: www.elsevier.com/locate/tvjl

Infectious diseases in large-scale cat hoarding investigations

K.C. Polak [a], J.K. Levy [a,*], P.C. Crawford [a], C.M. Leutenegger [b], K.A. Moriello [c]

- 4 large-scale hoarding operations
  - > 2,000 cats (Nov 2009 – March 2012)
  - IDEXX Feline Diarrhea RealPCR Panel
  - 68 cats with diarrhea

Table 2
Enteropathogens identified by PCR testing in 68 cats with diarrhea.

	FPV	FCoV	*C. jejuni*	*C. perfringens*	*C. coli*	*Salmonella* spp.	*T. foetus*	*Cryptosporidium* spp.	*Giardia* spp.	*T. gondii*
Case 1	0% (0/17)	82% (14/17)	–	0% (0/17)	–	0% (0/17)	35% (6/17)	12% (2/17)	47% (8/17)	0% (0/17)
Case 2	0% (0/17)	100% (17/17)	–	35% (6/17)	–	0% (0/17)	47% (8/17)	12% (2/17)	82% (14/17)	0% (0/17)
Case 3	0% (0/10)	90% (9/10)	–	60% (6/10)	–	0% (0/10)	50% (5/10)	20% (2/10)	80% (8/10)	0% (0/10)
Case 4	0% (0/24)	83% (20/24)	50% (12/24)	88% (21/24)	4% (1/24)	0% (0/24)	29% (7/24)	8% (2/24)	29% (7/24)	0% (0/24)
Total	0% (0/68)	88% (60/68)	50% (12/24)	49% (33/68)	4% (1/24)	0% (0/68)	39% (27/68)	12% (8/68)	56% (38/68)	0% (0/68)

FPV, feline panleukopenia virus; FCoV, feline coronavirus

Enteropathogens prevalence only

Paris et al. BMC Veterinary Research 2014, 10:13
http://www.biomedcentral.com/1746-6148/10/13

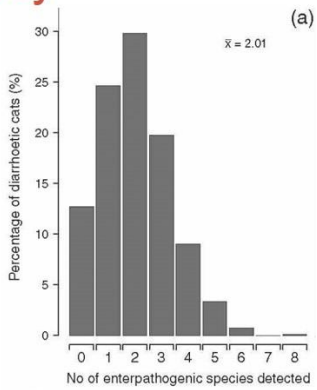
BMC Veterinary Research

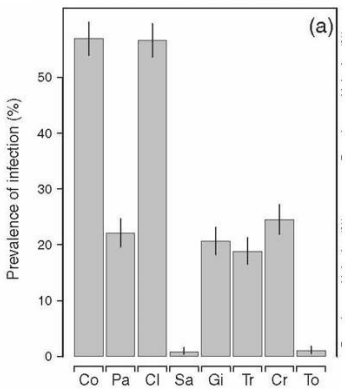
RESEARCH ARTICLE Open Access

Enteropathogen co-infection in UK cats with diarrhoea

Jasmin K Paris[1*], Sheila Wills[2], Hans-Jörg Balzer[3], Darren J Shaw[1] and Danièlle A Gunn-Moore[1]

- U.K. cats with diarrhea
  - 1,151 cats (Jun 2010 – Jan 2012)
  - IDEXX Feline Diarrhea RealPCR Panel
  - Multiple co-infections are common (62.5%)
  - No enteropathogen identified (12.7%)
  - *T. foetus* was co-associated with feline coronavirus, *C. perfringens* and *Giardia*





PCR panels

diarrhea –vs- no diarrhea

Enteropathogens identified in cats entering a Florida animal shelter with normal feces or diarrhea

Stephanie J. Sabshin, DVM; Julie K. Levy, DVM, PhD, DACVIM; Tiffany Tupler, DVM; Sylvia J. Tucker, BS; Ellis C. Greiner, PhD; Christian M. Leutenegger, DVM, PhD

Table 3—Frequency of identification of specific enteropathogens in fecal samples from the cats in Table 1.

Enteropathogen	Fecal consistency	No. tested	No. (%) positive	OR	95% CI	*P* value
Cryptosporidium spp	Normal	50	10 (20)	Referent		
	Diarrhea	50	5 (10)	0.44	NA	0.16
Cystoisospora spp	Normal	50	5 (10)	Referent		
	Diarrhea	50	7 (14)	1.47	NA	0.54
Giardia spp	Normal	50	4 (8)	Referent		
	Diarrhea	50	10 (20)	2.88	NA	0.08
Toxoplasma gondii	Normal	50	0 (0)	NA		
	Diarrhea	50	0 (0)	NA	NA	NA
Tritrichomonas foetus	Normal	50	0 (0)	NA		
	Diarrhea	50	0 (0)	NA	NA	NA
Clostridium perfringens enterotoxin A	Normal	50	25 (50)	Referent		
	Diarrhea	50	21 (42)	0.72	0.30–1.72	0.42
Salmonella spp	Normal	50	2 (4)	Referent		
	Diarrhea	50	3 (6)	1.53	NA	1.00
Ascarids	Normal	50	8 (16)	Referent		
	Diarrhea	50	3 (6)	0.34	NA	0.11
Hookworms	Normal	50	9 (18)	Referent		
	Diarrhea	50	5 (10)	0.44	0.16–1.19	0.07
Spirometra mansonoides	Normal	50	1 (2)	Referent		
	Diarrhea	50	0 (0)	NA	NA	1.00
Astrovirus	Normal	50	1 (2)	Referent		
	Diarrhea	50	4 (8)	4.26	NA	0.36
Calicivirus	Normal	50	1 (2)	Referent		
	Diarrhea	50	0 (0)	NA	NA	1.00
Feline coronavirus	Normal	50	18 (36)	Referent		
	Diarrhea	50	29 (58)	2.46	1.02–5.97	0.03
FPV	Normal	50	2 (4)	Referent		
	Diarrhea	50	2 (4)	1.00	NA	1.00

Bacterial enteropathogens

diarrhea –vs- no diarrhea

J Vet Intern Med 2012;26:54–60

Prevalence of Selected Bacterial and Parasitic Agents in Feces from Diarrheic and Healthy Control Cats from Northern California

E.V. Queen, S.L. Marks, and T.B. Farver

Table 1. Prevalence of bacterial enteropathogens in diarrheic and apparently healthy cats.

Organism	Diarrheic	Nondiarrheic	*P* Value
Campylobacter spp. (culture)	9.6% (21/219)	27.8% (15/54)	.001
Clostridium perfringens (culture)	42.8% (92/215)	63.0% (34/54)	.009
C. difficile (culture)	1.4% (3/215)	0% (0/54)	1
C. perfringens enterotoxin	4.1% (9/219)	1.9% (1/54)	.69
C. difficile toxin A	0% (0/219)	0% (0/54)	1
Pleisiomonas shigelloides	2.7% (6/219)	5.6% (3/54)	.39

PONAZURIL FOR COCCIDIOSIS

Sulfadimethoxine (Albon) is the only drug approved for treatment of coccidiosis in dogs and cats. This drug is coccidiostatic and requires a prolonged duration of administration. Recently the use of ponazuril (toltrazuril sulfone) has gained popularity for treatment of coccidiosis in dogs and cats. Ponazuril is available in the U.S. in paste form (Marquis paste®, Bayer Animal Health) as a treatment for *Sarcocystis neurona* in horses. A recent study examined the efficacy of treatment with ponazuril paste at each of three dosages in shelter-housed dogs and cats with confirmed coccidiosis. Dogs and cats treated with 50 mg/kg q 24 h for 3 days showed the greatest reduction in oocyst counts. The treatment protocol was associated with a 92.2% clearance of infection in dogs and 87.5% in cats. Animals with high pre-treatment oocyst counts were more likely to remain infected, many of which cleared the infection with a second treatment course. Due to prolonged infectivity of *Cystoisospora* spp. oocysts, environmental decontamination requires contaminated surfaces to be left in contact with 10% ammonia for at least 10 min or steam cleaned. Bathing infected animals may reduce oocyst contamination of the haircoat.

For dilution of Marquis paste, add 20 mL of water to 10 ml (=10 mg) of Marquis paste (15% ponazuril) to achieve a final solution of approximately 50 mg/ml. Mixture does not need to be refrigerated. Shake well before dosing; dispose of after 30 days. Alternatively ponazuril can be commercially compounded.

REFERENCES

1. U.S. Pet Ownership and Demographics Sourcebook. 2007.
2. Mott M. US faces growing feral cat problem. *National Geographic News* 2004.
3. www.feralcatproject.org. Accessed February 2010.
4. Scott FW, Geissinger, C. Kitten Mortality Survey. *Feline Practice* 1978;8:31-34.
5. Jemmett JE, Evans, J.M. A survey of sexual behavior and reproduction in female cats. *J Small Anim Pract* 1977;18:31-37.
6. van Aarde RJ. Population biology and control of feral cats on Marion island. *Acta Zool Fennica* 1984;172:107-110.

7. Nutter FB, Levine JF, Stoskopf MK. Reproductive capacity of free-roaming domestic cats and kitten survival rate. *J Am Vet Med Assoc* 2004;225:1399-1402.
8. Young C. Preweaning mortality in specific pathogen-free cats. *J Small Anim Pract* 1973;14:391-397.
9. Sparkes AH, Rogers K, Henley WE, et al. A questionnaire-based study of gestation, parturition and neonatal mortality in pedigree breeding cats in the UK. *J Feline Med Surg* 2006;8:145-157.
10. Root MV, Johnston SD, Olson PN. Estrous length, pregnancy rate, gestation and parturition lengths, litter size, and juvenile mortality in the domestic cat. *J Am Anim Hosp Assoc* 1995;31:429-433.
11. Strom Holst B, Frossling J. The Swedish breeding cat: population description, infectious diseases and reproductive performance evaluated by a questionnaire. *J Feline Med Surg* 2009;11:793-802.
12. Personal communication - Jim Babbitt, Chief Veterinarian San Diego Humane Society and SPCA.
13. Personal communication - Mondy Lamb, Marketing Director, SPCA of Wake County.
14. Cave TA, Thompson H, Reid SW, et al. Kitten mortality in the United Kingdom: a retrospective analysis of 274 histopathological examinations (1986 to 2000). *Vet Rec* 2002;151:497-501.
15. Bucheler J. Fading kitten syndrome and neonatal isoerythrolysis. *Vet Clin North Am Small Anim Pract* 1999;29:853-870, v.
16. Lawler DF. Investigating kitten deaths in catteries. *Consultations in Feline Internal Medicine* 1991:47-54.
17. Levy JK, Crawford PC, Collante WR, et al. Use of adult cat serum to correct failure of passive transfer in kittens. *J Am Vet Med Assoc* 2001;219:1401-1405.
18. Crawford PC, Levy JK, Werner LL. Evaluation of surrogate markers for passive transfer of immunity in kittens. *J Am Vet Med Assoc* 2006;228:1038-1041.
19. Murray JK, Skillings E, Gruffydd-Jones TJ. A study of risk factors for cat mortality in adoption centres of a UK cat charity. *J Feline Med Surg* 2008;10:338-345.