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Infectious diseases in large-scale cat hoarding investigations

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ABSTRACT

Animal hoarders accumulate animals in over-crowded conditions without adequate nutrition, sanitation, and veterinary care. As a result, animals rescued from hoarding frequently have a variety of medical conditions including respiratory infections, gastrointestinal disease, parasitism, malnutrition, and other evidence of neglect. The purpose of this study was to characterize the infectious diseases carried by clinically affected cats and to determine the prevalence of retroviral infections among cats in large-scale cat hoarding investigations. Records were reviewed retrospectively from four large-scale seizures of cats from failed sanctuaries from November 2009 through March 2012. The number of cats seized in each case ranged from 387 to 697. Cats were screened for feline leukemia virus (FeLV) and feline immunodeficiency virus (FIV) in all four cases and for dermatophytosis in one case. A subset of cats exhibiting signs of upper respiratory disease or diarrhea had been tested for infections by PCR and fecal flotation for treatment planning.

Mycoplasma felis (78%), calicivirus (78%), and *Streptococcus equi* subspecies *zooepidemicus* (55%) were the most common respiratory infections. Feline enteric coronavirus (88%), *Giardia* (56%), *Clostridium perfringens* (49%), and *Tritrichomonas foetus* (39%) were most common in cats with diarrhea. The seroprevalence of FeLV and FIV were 8% and 8%, respectively. In the one case in which cats with lesions suspicious for dermatophytosis were cultured for *Microsporum canis*, 69/76 lesional cats were culture-positive; of these, half were believed to be truly infected and half were believed to be fomite carriers. Cats from large-scale hoarding cases had high risk for enteric and respiratory infections, retroviruses, and dermatophytosis. Case responders should be prepared for mass treatment of infectious diseases and should implement protocols to prevent transmission of feline or zoonotic infections during the emergency response and when transferring the rescued cats to other shelters or to adopters.

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Introduction

Animal hoarding is a complex human and animal welfare issue that exists in almost every community. Hoarding cases can involve dozens to hundreds of animals, dead and alive, living in squalid conditions (Patronek, 2001). Suspected hoarding cases frequently come to public attention when individuals amass large numbers of animals with good intentions, but lack capacity for providing minimal standards of care (Lockwood, 1994).

While the true prevalence of hoarding is unknown due to the social isolation of hoarders, dismissive responses by potential reporters, and a lack of legal investigative authority or motivation to pursue hoarding cases, an estimated 700–2000 new cases are re-

ported per year (Patronek, 1999). Hoarding occurs among individuals as well as those operating animal shelters, sanctuaries, and rescue groups (Miller and Zawistowski, 2012).

Hoarding conditions are often characterized by an accumulation of animal waste, carcasses, elevated ammonia levels, and garbage, leading to compromised animal welfare and health (Campbell and Robinson, 2001). Animals can be found in over-crowded conditions without provision for adequate food, water, sanitation and veterinary care. As a result, animals seized from hoarding situations frequently suffer from a variety of medical conditions including respiratory infections, diarrhea, parasitism, and skin diseases (Patronek, 2008; Reinisch, 2009).

Legal and humane interventions in large-scale cases can be procedurally cumbersome and costly, often requiring a multi-faceted response involving representatives from animal control, humane societies, veterinary medicine, public health, code enforcement, social services, and law enforcement (Patronek, 2001). When the number

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of animals and the severity of conditions exceed the response capacity of local agencies, a collaborative effort between local and national agencies is often required (Lockwood, 1994).

Responses to suspected large-scale cruelty cases often consist of both forensic and animal rescue components. During the animal rescue, animals are often triaged onsite and then transported to a temporary shelter to await their legal disposition. If the owner loses legal custody of the animals, they might be adopted, transferred to other agencies, or euthanased. Historically, the high prevalence of infectious diseases and lack of socialization of cats seized in large-scale cases often resulted in mass euthanasia in shelters already crowded by pet overpopulation.^{1,2} In recent years, responding agencies have invested in the physical and behavioral rehabilitation of seized animals to reduce their euthanasia.³ This has increased the number of animals eventually placed in homes and the cost of intervening in suspected cruelty cases. The costs have exceeded \$1–2 million⁴ in the largest cases.^{5,6}

Medical conditions complicate the long-term sheltering and adoption of seized animals. Infectious diseases pose a risk to other animals in the shelter and potentially to pets in the community. Some infections could have zoonotic risk to both shelter staff and potential adopters. Knowledge of conditions commonly present in large-scale cruelty investigations would assist with planning for the care of seized animals, including ordering of medical supplies, preparation of shelter protocols, and eventual placement of the animals. Although media reports frequently cite high disease rates from such cases, there are no detailed descriptions of diseases diagnosed in cats seized in cruelty investigations.^{5,7}

From November 2009 through March 2012, more than 2000 cats were removed from four cat sanctuaries following reports consistent with animal hoarding. In each case, veterinarians performed intake examinations and screened for infectious diseases as needed to guide treatment decisions for ill cats and to segregate infectious cats housed in temporary shelters. The purpose of this study was to examine the available records of cats removed during the four large-scale cruelty investigations to characterize the infectious diseases of cats with respiratory and gastrointestinal signs and to determine the prevalence of retroviral infections.

Materials and methods

Cat sanctuaries

Case 1

In November 2009, 594 cats were relinquished from a cat sanctuary in Florida following a site assessment that revealed high rates of disease and mortality. The majority of cats were group-housed in indoor/outdoor wire mesh pens. Cats under treatment were housed in a barn in plastic airline carriers. Following relinquishment, cats were examined and sheltered on-site for several weeks pending disposition.

Case 2

In June 2010, 387 cats were relinquished from a Pennsylvania cat sanctuary after an inspection revealed the cats to be housed in overcrowded and unsanitary conditions on the first floor of a two-story commercial building. The majority of cats were group-housed indoors. Following relinquishment, cats were transported to a

temporary shelter established at a nearby fairground for intake examinations. There they were housed in stacked wire crates for several weeks pending disposition.

Case 3

In June 2011, 697 cats were seized from a cat sanctuary in Florida. Cats were group-housed in outdoor community pens, barn pens, travel trailers, or indoor wire crates. Cats were transported to several air-conditioned warehouses where intake examinations were performed. Cats were housed in stacked wire cages in the temporary shelter for approximately 3 months until disposition.

Case 4

In February 2012, 696 cats were seized from a Florida cat sanctuary. The majority of cats roamed freely outdoors, with a few dozen housed in pens or trailers designated for diseased animals. Movement of cats among the various housing areas was unrestricted. Cats were triaged in the field and those in critical condition were taken to animal hospitals for care. Cats healthy enough for transport were relocated to a vacant animal shelter where they received intake examinations. The cats were then group-housed two to four per indoor/outdoor run or in individual cages until disposition approximately 6 months post-seizure.

Intake examinations

Intake examinations were performed on all cats on-site at the sanctuary (case 1) or upon arrival at the temporary shelters (cases 2–4). Intake protocols varied between the four cases but all included a physical examination that documented each cat's estimated age, breed, sex, weight, and body condition score. The presence of illness or injuries was recorded.

Cats were photographed, vaccinated, treated for internal and external parasites, and blood was collected for retroviral testing. Specimens were collected from a subset of the cats exhibiting signs of respiratory (ocular or nasal discharge, sneezing, coughing, conjunctivitis, or blepharospasm) or gastrointestinal (diarrhea) disease. The criteria for performing diagnostic testing varied between cases and were based on the participating agency's medical protocols and the decisions made by the supervising veterinarians.

Respiratory specimens were collected at the time of intake examination. Fecal specimens were usually collected during the first 5 days of housing at the temporary shelters when diarrhea was observed. A small subset of 15 cats from case 4 were sampled 14 days post-intake to the temporary shelter due to persistence of diarrhea despite anti-parasite treatments (pyrantel pamoate, praziquantel, ponazuril) administered during the intake examination.

Serological testing

In all cases, cats were tested for feline leukemia virus (FeLV) antigen and feline immunodeficiency virus (FIV) antibody with a commercially available ELISA (SNAP Feline Triple Test or SNAP Combo Test, IDEXX Laboratories) using whole blood. Testing for *D. immitis* antigen (SNAP Feline Triple Test, IDEXX Laboratories) was performed in the majority of cats 7 months and older in two of the four cases.

Testing of cats with respiratory disease

Diagnostic specimens from cats with signs of respiratory disease were collected by swabbing the conjunctiva for PCR detection of pathogens. One specimen was collected by rolling a dry polyester swab (Fisherbrand, Thermo Fischer Scientific) in the ventral conjunctival sac and a second swab was collected by rubbing a dry swab around the oropharynx. The paired swabs from each cat were placed together in a sterile dry polystyrene vial for pooled analysis and labeled with pertinent specimen identification information such as specimen collection date, animal identification, and agency name. Fresh latex gloves were worn during specimen collection for each cat and the vial containing the paired swabs was placed in an individual plastic self-sealing bag to reduce cross-contamination prior to testing. The specimens were stored at 4 °C pending analysis within 72 h of collection by a commercial reference laboratory (Feline Upper Respiratory Disease RealPCR Panel, IDEXX Laboratories).

Respiratory specimens were tested by real-time PCR for feline herpesvirus type 1 (FHV) glycoprotein B (*gB*) gene, S66371; feline calicivirus (FCV) ORF 1 gene, AF109465; influenza virus H₁N₁ type A influenza hemagglutinin HA gene, GQ229373; *Streptococcus equi* subspecies *zooepidemicus* (SEZ) AroA, 3-phosphoshikimate 1-carboxyvinyltransferase gene, FM204884; *Bordetella bronchiseptica* hemagglutinin fusion protein gene (*FhaB*), AF140678; *Mycoplasma felis* single-stranded rRNA-internal transcribed region 1 (ITS-1) gene, AF443608; and *Chlamydomphila felis* outer membrane protein A (*OmpA*) gene, AP00686. Real-time PCR was performed with seven quality controls, including PCR-positive controls, PCR-negative controls, negative extraction controls, DNA pre-analytic quality control targeting the host 18S rRNA gene complex, RNA pre-analytic quality control targeting the host 18S rRNA gene complex,

¹ See: <http://www.examiner.com/article/85-pets-removed-from-hoarder-70-euthanized> (accessed 14 May 2014).

² See: http://www.wyomingnews.com/articles/2010/02/08/featured_story/01top_02-08-10.txt (accessed 14 May 2014).

³ See: <http://www.aspc.org/about-us/press-releases/aspc-opens%20behavioral-rehabilitation-center-help-animal-victims-cruelty> (accessed 14 May 2014).

⁴ \$1 = approx. £0.59; €0.73 as at 17 May 2014.

⁵ See: <http://www.gainesville.com/article/20120329/ARTICLES/120329495> (accessed 14 May 2014).

⁶ See: http://www.huffingtonpost.com/ed-sayres/hundreds-of-caboodle-ranc_b_1747467.html (accessed 14 May 2014).

⁷ See: <http://www.jsonline.com/199430911.html> (accessed 14 May 2014).

Table 1
Pathogens identified by PCR testing in 81 cats with clinical signs of respiratory disease.

	FCV	FHV	H ₁ N ₁	SEZ	<i>B. bronchiseptica</i>	<i>M. felis</i>	<i>C. felis</i>
Case 1	45% (9/20)	35% (7/20)	–	13% (4/31)	20% (4/20)	80% (16/20)	0% (0/20)
Case 2	72% (18/25)	12% (3/25)	0% (0/25)	76% (19/25)	28% (7/25)	76% (19/25)	60% (15/25)
Case 3	100% (9/9)	2% (2/9)	0% (0/9)	33% (3/9)	11% (1/9)	56% (5/9)	56% (5/9)
Case 4	96% (27/27)	41% (15/27)	0% (0/27)	93% (25/27)	18% (2/27)	81% (21/27)	5% (1/27)
Total	78% (63/81)	33% (27/81)	0% (0/81)	55% (51/92)	17% (14/81)	78% (61/81)	26% (21/81)

FCV, feline calicivirus; FHV, feline herpesvirus; H₁N₁, influenza virus H₁N₁; SEZ, *Streptococcus equi* subspecies *zoepidemicus*.

an internal positive control spiked into the lysis solution, and an environmental contamination monitoring control.

Testing of cats with diarrhea

Fecal specimens were collected from the litter box, cage floor, or by rectal swab from cats with diarrhea. To avoid cross-contamination of specimens, each fecal specimen was collected using fresh gloves and assisted by a fresh wooden tongue depressor or fresh syringe depending on the consistency of the specimen. Fecal aliquots were split into two fresh plastic screw-top jars. One jar from each specimen was selected for PCR assay and was individually placed into its own plastic self-sealing bag to further reduce the chance of cross-contamination. The second jar was designated for fecal flotation. All supplies were disposed of after each specimen and were not re-used. Specimens were stored at 4 °C pending analysis within 72 h of collection at the reference laboratory (IDEXX Feline Diarrhea RealPCR Panel, IDEXX Laboratories).

Fecal specimens were tested by real-time PCR for a panel of potential enteropathogens, including feline panleukopenia virus (FPV) VP2 gene, EU252145; feline coronavirus (FCoV) 7b gene, DQ010921.1; *Campylobacter jejuni* UDP-N-acetylglucosamine acyltransferase gene *lpxA*, AL111168; *Clostridium perfringens* alpha toxin (CPA) gene, phospholipase C, L43545; *Campylobacter coli* UDP-N-acetylglucosamine acyltransferase *lpxA* gene, AY531496; *Salmonella* spp. invasion A gene, EU348366; *Tritrichomonas foetus* 5.8S rRNA gene, AF339736; *Cryptosporidium* spp. small-subunit rRNA gene, A093489; *Giardia* spp. small-subunit rRNA gene, DQ836339; and *Toxoplasma gondii* internal transcribed spacer-1 gene, L49390. The same quality controls were performed as the respiratory PCR. Fecal specimens were also tested by zinc sulfate centrifugation to detect parasite cysts, oocytes, and larvae at a reference laboratory (IDEXX Reference Laboratories).

Dermatophyte screening

Undercover investigators suspected dermatophytosis to be widespread in case 4 prior to seizure of the cats, so the cats were screened for clinically obvious signs of infection at the time of the intake examination at the temporary shelter. Each cat was assessed in room light for skin lesions and in a dark room with a Wood's lamp to screen for fluorescing hairs. Specimens were collected using a freshly unsealed toothbrush from all cats with skin lesions or cats that had fluorescing hairs on Wood's lamp examination. Suspicious areas were brushed at least 30 times. Specimens were transported via overnight courier with ice packs to a university laboratory (University of Wisconsin, School of Veterinary Medicine, Dermatology Research Laboratory) for testing. The specimens were plated onto dermatophyte culture media in Petri dish plates (90 × 15 mm) within 72 h of collection (Mycosel agar modified with phenol red as a color indicator, Becton Dickinson), incubated at 27–30 °C for 21 days, and examined daily for growth.

Results

Cats

No original records from the cat sanctuaries were available, so information for this report was collected retrospectively from the examination records completed at the time cats were relinquished

or seized. The vast majority of cats in each case were adults. Of cats for which estimated ages were available, 98% (1956/1989) were estimated to be >6 months of age. Of cats for which gender information was available, 52% were female (939/1790).

Respiratory infections

Signs of upper respiratory infection (URI) were observed during intake examinations in 27% (366/1368) of cats for which records of physical findings were available. The proportion of cats in each case with signs of URI ranged from 16% to 38%.

A subset of 81 cats with respiratory signs was tested for respiratory infections (Table 1). FCV (78%), *M. felis* (78%), and SEZ (55%) were the most prevalent infectious agents, followed by FHV (33%), *C. felis* (26%), and *B. bronchiseptica* (17%). H₁N₁ influenza was not identified in any cat. The majority of tested cats were infected with multiple pathogens (Table 4). In cats in which SEZ was identified, 88% (45/51) and 78% (40/51) of cats were co-infected with FCV and *M. felis*, respectively.

Enteric infections

A total of 68 diarrheic fecal specimens were submitted for PCR testing (Table 2). The most commonly identified enteropathogens included FCoV (88%), *Giardia* spp. (56%), *C. perfringens* (49%), *C. jejuni* (50%) and *T. foetus* (39%). FPV, *Salmonella* spp., and *T. gondii* were not detected in any specimens. A total of 95 fecal specimens were tested for parasites using zinc sulfate centrifugation (Table 3). Hookworms were identified in the sanctuaries in which cats had contact with earth (cases 3 and 4), whereas ascarids were identified in all four cases. Most tested specimens harbored multiple enteropathogens (Table 4). In cats infected with *T. foetus*, 67% were co-infected with *Giardia* spp.

FeLV and FIV infections

In the three sanctuaries from which results from FeLV/FIV testing were available for review (cases 1, 3 and 4), the overall seroprevalences of FeLV and FIV were 8% and 8%, respectively (Table 5). A total of 1% of cats were positive for both FeLV and FIV. In all four cases, retroviral-infected cats were found to be roaming freely with uninfected cats and were not restricted to areas formally designated for segregating infected cats.

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

	FPV	FCoV	<i>C. jejuni</i>	<i>C. perfringens</i>	<i>C. coli</i>	<i>Salmonella</i> spp.	<i>T. foetus</i>	<i>Cryptosporidium</i> spp.	<i>Giardia</i> spp.	<i>T. gondii</i>
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.

Table 3
Enteropathogens identified in 95 cats with diarrhea by zinc sulfate centrifugation.

	Ascarids	Whipworms	Cystoisospora	Hookworms	Nematodirus
Case 1	19% (3/17)	0% (0/17)	0% (0/17)	0% (0/17)	0% (0/17)
Case 2	6% (1/17)	0% (0/17)	6% (1/17)	0% (0/17)	6% (1/17)
Case 3	53% (8/15)	0% (0/15)	13% (2/15)	47% (7/15)	0% (0/15)
Case 4	2% (1/46)	7% (3/46)	0% (0/46)	4% (2/46)	0% (0/46)
Total	14% (13/95)	3% (3/95)	3% (3/95)	9% (9/95)	1% (1/95)

Table 4
Frequency of identification of multi-pathogen co-infections in individual cats with respiratory disease or diarrhea.

Number of pathogens per cat	0	1	2	3	4	5
Respiratory pathogens	3% (2/81)	9% (7/81)	17% (14/81)	42% (34/81)	26% (21/81)	4% (3/81)
Enteropathogens	0% (0/68)	15% (10/68)	29% (20/68)	29% (20/68)	18% (12/68)	7% (5/68)

Heartworm infection

Only 1/1048 cats tested for *Dirofilaria immitis* antigen had a positive test.

Dermatophyte infections

Finances did not allow for fungal culture of all of the cats in case 4 and culturing was limited to cats with a high degree of suspicion for dermatophytosis based on the presence of suspect skin lesions and/or Wood's lamp examination. All cats received one lime sulfur dip at intake pending fungal culture results.

Of the total population of 696 cats, it was suspected that 76 cats were infected; of these, 69 (91%) were culture-positive for *M. canis*. A total of 14/69 culture-positive cats had <9 colony forming units per plate (cfu/plate); of these, all had skin lesions but only one had a positive Wood's lamp examination. All cats with confirmed dermatophyte infections were treated with twice weekly lime-sulfur dips at a 1:16 dilution and oral terbinafine at 35 mg/kg once daily until they had two consecutive negative weekly fungal cultures.

Discussion

Symptomatic cats from all four large-scale cruelty investigations were found to carry a variety of infectious diseases. This was not unexpected given the large number of cats confined in crowded conditions and the failure to isolate diseased cats from clinically normal cats.

Clinical signs of respiratory disease were observed in cats rescued from all four cases. In this study, the most prevalent respiratory pathogens were *M. felis*, FCV, and SEZ. All three pathogens have been reported in cats both with and without clinical signs of disease, making segregation of populations based on clinical signs alone an ineffective biosecurity strategy (McManus et al., 2014). The role that *M. felis* and SEZ play in feline respiratory disease is currently poorly defined. They may act as primary pathogens or exert their effects only in combination with other infectious co-factors (Randolph et al., 1993). *Mycoplasma* infection has been signifi-

Table 5
Retroviral infections identified in 1940 cats in three cases.

	FeLV	FIV
Case 1	9% (52/557)	7% (40/557)
Case 3	8% (53/689)	3% (22/689)
Case 4	7% (48/694)	13% (87/694)
Total	8% (153/1940)	8% (149/1940)

FeLV, feline leukemia virus; FIV, feline immunodeficiency virus.

cantly associated with feline URI in animal shelters, but most affected cats also carried co-infections such as FHV (Bannasch and Foley, 2005; Veir et al., 2008; Hartmann et al., 2010; Holst et al., 2010; Burns et al., 2011; Litster et al., 2012). *M. felis*-infected shelter cats treated with a 14-day course of oral doxycycline had clinical improvement, although clearance of the organism was not demonstrated (Kompare et al., 2012).

FCV was the most common viral respiratory pathogen detected (78%), surpassing the prevalence rate of FHV (33%). FHV persists as a latent infection in cats and can recrudescence during periods of stress (Gaskell et al., 2007). As a consequence of stress and cross-contamination, FHV is the most prevalent viral pathogen in cats with acute URI in traditional short-term shelter housing (Bannasch and Foley, 2005; Veir et al., 2008; McManus et al., 2014). In contrast to cats in short-term shelters, foster programs, or trap–neuter–return programs, cats housed long-term in sanctuaries are more likely to be infected with FCV than FHV, similar to the findings of cats from the cases in this report (McManus et al., 2014). The high prevalence of FCV among cats with URI is concerning because of considerable strain variability in its pathogenicity, transmissibility, and environmental stability and its association with a wide array of clinical signs including fever, conjunctivitis, oral ulceration, limping, ocular and nasal discharge, and gingivostomatitis (Reubel et al., 1992; Dawson et al., 1994; Pedersen et al., 2000; Radford et al., 2007; Gerriets et al., 2012). The reason for the shift from high prevalence of FHV in cats in short-term shelter housing to high prevalence of FCV in cats in long-term group-housed confinement is unknown (Reubel et al., 1992; Lommer and Verstraete, 2003; Dowers et al., 2010).

SEZ was detected with high frequency in cats with respiratory disease. SEZ is considered to be an emerging pathogen in animal shelters and has been reported as the causative agent of several outbreaks of fatal hemorrhagic pneumonia disease in dogs (Garnett et al., 1982; Chalker et al., 2003; Kim et al., 2007; Pesavento et al., 2008; Byun et al., 2009; Priestnall and Erles, 2011). SEZ can also be found in asymptomatic dogs, and it is possible that infectious co-factors such as canine distemper virus, canine influenza virus, or canine pneumovirus are required for maximum virulence (Byun et al., 2009; Larson et al., 2011).

In cats, SEZ infections are associated with purulent nasal discharge, dyspnea, coughing, pneumonia, rhinitis, meningoencephalitis, and death, but the overall prevalence of SEZ in sheltered cats has yet to be determined (Britton and Davies, 2010; Martin-Vaquero et al., 2011). SEZ was reported as the causative agent of death in 78 cats from June 2006 to January 2008 in an Israeli cattery (Blum et al., 2010). The common finding of SEZ in cats in large-scale cases could explain in part why URI appears to be more severe and even deadly in such circumstances, in contrast to the milder and self-limiting

URI commonly associated with cats in short-term shelters in which SEZ does not appear to be commonly involved. SEZ is likely an under-recognized cause of severe disease in cats since screening is not currently included in routine laboratory test panels, and the clinical presentation is different to the dramatic hemorrhagic respiratory disease observed in dogs.

B. bronchiseptica is a Gram-negative bacterium considered to be a primary respiratory pathogen of cats found most commonly in high-density housing environments such as shelters or sanctuaries (Helps et al., 2005; Egberink et al., 2009). Clinical signs vary in severity from mild sneezing and ocular discharge to pneumonia and death (Willoughby et al., 1991; Welsh, 1996; Speakman et al., 1999). *C. felis* is currently recognized as primarily an ocular pathogen causing conjunctivitis but continues to be isolated from cats with respiratory tract disease. Reported infection rates range from 1% to 5% of cats without signs of URI to 10–30% of cats with conjunctivitis or URI (Wills et al., 1988; Gruffydd-Jones et al., 1995; Sykes et al., 1999; Low et al., 2007; McManus et al., 2014).

A total of 14 different potential enteropathogens were identified in cats with diarrhea, including four protozoan pathogens. *T. foetus* was detected with high frequency in cats from all four cases, with prevalences ranging from 29% to 50% of cats tested. *T. foetus* is a highly contagious causative agent of intermittent large bowel diarrhea in cats, particularly when cats are confined in group-housing (Gookin et al., 1999, 2004; Foster et al., 2004; Holliday et al., 2009; Tolbert and Gookin, 2009). The diagnosis, management, and adoption of cats infected with *T. foetus* can be difficult in a shelter setting. *T. foetus* is often not identified, or misdiagnosed as *Giardia* spp. on fecal flotation, which is an important consideration because 67% of cats infected with *T. foetus* in this study were co-infected with *Giardia* spp.

Currently there is no approved treatment for feline tritrichomoniasis, and response to therapy is variable. The antiprotozoal drug ronidazole is frequently used but can cause reversible neurotoxicosis (Gookin et al., 2006). Due to the high potential for transmission, infected cats should be immediately isolated and treated with ronidazole administered at the recommended dose of 30 mg/kg daily for 14 days. PCR testing should be repeated after treatment is finished to assure treatment success. Regardless of testing results, however, cats should be kept isolated until after their adoption to minimize spread to other cats as relapse of infection can occur as long as 20 weeks post-treatment.

Giardia spp. was identified in 56% of fecal specimens tested. This exceeds prevalences previously reported in sheltered cats both with and without diarrhea in New York (8%), California (10%), or Florida (20%; Spain et al., 2001; Mekaru et al., 2007; Sabshin et al., 2012; McManus et al., 2014). *Cryptosporidium* spp. was detected in all four cases at higher prevalences than previously reported in sheltered cats both with and without diarrhea from Colorado (8%), New York (5%), Florida (20%; Hill et al., 2000; Spain et al., 2001; Sabshin et al., 2012; McManus et al., 2014). *Cystoisopora* spp. was infrequently detected, which is most likely because of the low proportions of kittens present in all cases (Gates and Nolan, 2009). The high prevalence of *Giardia* and *Cryptosporidium* spp. can be attributed to the increased population density and continuous reinfection of cats from accumulated feces found in hoarding cases. Both pathogens create management challenges due to their environmental durability, potential for misdiagnosis, and lack of practical curative treatment options.

Nematodes with zoonotic potential, ascarids and hookworms, were identified in 14% and 9% of symptomatic cats tested, respectively. Previous studies have identified the prevalence of hookworms in outdoor-dwelling feral cats in Florida (75%), cats housed in a Florida shelter (14%), and in pet cats in Pennsylvania (8%; Anderson et al., 2003; Gates and Nolan, 2009; Sabshin et al., 2012). The collection of fecal specimens after intake parasite treatments

were administered might explain why the prevalence of ascarids detected in this study was much lower than prevalence rates reported in northern, mid-Atlantic and central states (Spain et al., 2001; Vasilopoulos et al., 2006).

One or more potentially pathogenic bacteria including *C. jejuni*, *C. coli*, and *C. perfringens* were detected in the majority of cats. However, the clinical significance of this is unclear, as *C. perfringens* has been identified in cats both with and without diarrhea (Sabshin et al., 2012). The quantity of CPA detected in feces might better correlate with diarrhea than its mere presence (Leutenegger et al., 2012).

The finding of FCoV in most cats is consistent with previous reports that documented higher seropositivity rates for FCoV antibodies in confined cats than in free-roaming stray and feral cats, presumably due to shared litter boxes (Pedersen, 2009). Infection with FCoV usually causes only a transient, mild diarrhea. Viral mutations can lead to fatal feline infectious peritonitis, which is anecdotally reported to be relatively common in hoarded cats (Pedersen, 2009).

Sanctuary managers in all four cases in this report described policies for all cats to be tested for FeLV and FIV and then segregated based on serological results. However, retroviral-infected cats were identified in each of the facilities, often group-housed with uninfected cats. The retroviral prevalence of cats from these cases surpasses infection rates reported for both pet and feral cats (Luria et al., 2004; Levy et al., 2006). This can be attributed to the increased physical contact, stress, and agonistic interactions between cats living in densely populated confinement. All cats should be tested for retroviruses at the time of seizure from hoarding cases as the retroviral status of cats will influence housing decisions, medical care, and adoption options. Cats should also be retested 2 months after the initial testing to identify early infections that escaped detection at the time of intake.

Dermatophytosis is an important consideration when sheltering cats from any source because it is highly contagious, has zoonotic potential, and can be difficult to diagnose and to treat (Carlotti et al., 2010). Its management is particularly challenging when sheltering large numbers of seized cats, because of the potential transmission of *Microsporium canis* in temporary shelters to both cats and to humans. Due to prior difficulties in managing dermatophytosis cases in temporary shelters, in case 4 all cats were dipped in lime-sulfur at intake to decrease the overall spore burden in the shelter and to reduce in-shelter transmission of disease. Cats that exhibited lesions suspected to be dermatophytosis were immediately segregated. Cats with skin lesions, Wood's lamp positive examinations, or culture-confirmed dermatophytosis were treated with terbinafine and lime-sulfur dips. This protocol was effective in eliminating infections in all treated cats.

In cruelty cases involving a limited number of cats, it is often practical to perform comprehensive diagnostic testing of each cat for infectious diseases and to develop individualized treatment plans based on test results. In contrast, large-scale cases involving hundreds of cats taken in and housed in makeshift quarters by emergency responders require population-level management. Medical protocols for both healthy and diseased cats must be developed prior to the seizure so they can be implemented at the time of first contact with the cats. Protocols used to date have been based primarily on the anecdotal experience of responding agencies.

While the actual proportion of cats tested in this retrospective study was low, the results provide useful information regarding the potential identity and prevalence of pathogens for which protocols can be tailored. The results reported in this study might not be representative of all cat hoarding cases, due to differences in geographical location, housing type, and level of care provided.

Similar to traditional animal shelters, at the time of intake examination pyrantel pamoate, ponazuril, praziquantal, and a topical treatment effective against fleas and mites should be adminis-

tered to all cats. All cats 4 weeks of age or older should be vaccinated against FPV, FCV, and FHV with a modified-live vaccine. If cats are to be co-housed, which is usually the case in large-scale operations, vaccination against FeLV should be initiated at intake to reduce the risk of continued transmission from cats that have infections that are not detected immediately (Levy et al., 2008). Cats 12 weeks of age or older should be vaccinated against rabies.

While antibiotic therapy should always be used judiciously to avoid the development of drug resistance, consideration should be given to the inclusion of antibiotics in the intake protocol due to the high prevalence and morbidity of SEZ in cats from large-scale cases. Cefovecin has been used effectively in the control of fatal SEZ outbreaks in shelter dogs in which a single dose provides broad-spectrum long-lasting activity and reduces handling stress, cross-contamination between animals, and labor costs. It is also an ideal empirical treatment for many commonly encountered conditions in cats seized from hoarding cases, including wounds, pyoderma, and otitis. Unfortunately, cefovecin is ineffective against other common bacterial pathogens encountered in cats with URI, such as *M. felis*, *B. bronchiseptica*, and *C. felis*. This means that effective treatment of cats with URI often requires combination therapy with other antibiotics such as doxycycline. Doxycycline should not be used alone for therapy however, as SEZ isolates are frequently resistant to doxycycline (Chalker et al., 2012).

The effort and expense of such broad-spectrum preventive health-care delivered concurrent with the physical examinations and forensic documentation should not be underestimated. However, it has been our experience that a well-planned and resourced intake procedure is more likely to result in comprehensive and consistent care than a less thorough protocol that relies on routine treatments to be provided at a later time.

Protocols should be in place for assessment of sick cats, including those identified at the time of intake as well as those that develop disease while in the temporary shelter. Diagnostic panels should include testing for bacterial, viral, and parasitic infections and include screening for pathogens unique to large-scale cases, such as SEZ. The diagnostic value of necropsy of cats found dead or euthanased should not be overlooked as a useful disease management tool. Diagnostic laboratories should be notified of the need for timely result reporting, particularly if highly pathogenic or unusual infections are detected.

Treatment protocols must be developed to ensure that cats receive consistent care, regardless of the personnel involved. Treatments must be practical and affordable within the context of large-scale cases, which are often staffed by rotating teams of emergency responders and volunteers. For example, mild to moderate URIs could be treated empirically with once daily doxycycline, whereas severe respiratory infections warrant diagnostic testing and more broad-spectrum coverage pending the results of testing.

Whether cats are discharged from temporary shelters directly to new adoptive homes or to another shelter for eventual placement, complete records of examination findings, diagnostic results, and treatments administered should be transmitted with each cat. The high carriage rate of persistent infections that are difficult to diagnose and to eliminate means that these cats pose a risk for transmission of new pathogens into any other facility they are transferred to. Therefore, cats rescued from hoarding cases should be segregated from any pre-existing cat shelter populations until they are rehomed.

Conclusions

This report is the first to describe infectious diseases detected in cats at the time of intervention in large-scale hoarding investigations. Most symptomatic cats harbored multiple respiratory and enteric pathogens, and the prevalence of retroviral infections was

higher than typically found in animal shelters. These findings can be used to guide preventive care protocols, but much more knowledge is needed to inform comprehensive management tactics for such cases, especially since hoarding is being reported with increasing frequency. Currently, intervention strategies tend to center on emergency management of individual situations, and there is little follow-up or transparency regarding case outcomes. Prospective research is needed to more fully characterize the infectious diseases present in both symptomatic and asymptomatic cats and the effectiveness of preventive care and treatment protocols. Additional topics deserving attention include the management of stress and pain in seized cats, the characterization and management of common noninfectious conditions, and the long-term outcomes of the cats. Case managers should prioritize these factors during incident response planning. This will enable the development of best-practice guidelines and cost:benefit analyses to guide responders across the industry and to continuously improve outcomes for rescued cats.

Conflict of interest statement

Dr. Christian Leutenegger is the Head of PCR at IDEXX Laboratories. None of the authors has any financial or personal relationships that could inappropriately influence or bias the content of the paper.

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