5-2-2024

Cytauxzoonosis in Indiana, USA: a case series of cats infected with Cytauxzoon felis (2018-2022)

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Recommended Citation

Reichard, M. V., Cotey, S. R., Dangoudoubiyam, S., Weerarathne, P., Tussey, K., Wilkes, R. P., & et. al. (2024). Cytauxzoonosis in Indiana, USA: a case series of cats infected with Cytauxzoon felis (2018-2022). *Journal of feline medicine and surgery, 26*(5), 1098612X231224139. http://doi.org/10.1177/1098612X231224139
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Case Series

jfms Journal of Feline Medicine and Surgery

Cytauxzoonosis in Indiana, USA: a case series of cats infected with *Cytauxzoon felis* (2018–2022)

Journal of Feline Medicine and Surgery
1–7

DOI: 10.1177/1098612X231224139
journals.sagepub.com/home/jfm

This paper was handled and processed by the American Editorial Office (AAFP) for publication in *JFMS*

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Abstract

Case series summary This case series describes six cases involving seven cats naturally infected with *Cytauxzoon felis* in Indiana, USA. Medical records were retrospectively reviewed and all available information on signalment, history, clinical and diagnostic findings, treatment, outcome and pathology was reported. Cats infected with *C felis* were domestic shorthairs, were aged between 2 and 9 years and all but one of the cats were male. The seven infected cats originated from five counties in southwestern Indiana. Six of seven cats were found to have acute cytauxzoonosis based on clinical signs, gross pathologic lesions, observation of *C felis* in tissues and/or detection of *C felis* DNA. One cat was identified as a subclinical survivor cat with no known clinical history of cytauxzoonosis. *Relevance and novel information* The reported cases are the first confirmed reports of acute and chronic cytauxzoonosis in cats from Indiana and document an expansion in the range of *C felis*. Veterinary practitioners in Indiana should consider infection with *C felis* as a differential diagnosis for cats that present with fever, inappetence, lethargy, depression, dehydration, dyspnea, hemolytic crisis, anorexia or icterus. Administration of approved acaricides to cats currently offers the best protection and control against *C felis* infection.

Keywords: *Cytauxzoon felis*; cytauxzoonosis; bobcat fever; Indiana; tick-borne disease; *Amblyomma americanum*; lone star tick; *Dermacentor variabilis*; American dog tick

Accepted: 8 December 2023

Introduction

Cytauxzoonosis, caused by infection with *Cytauxzoon felis*, is a severe and often fatal tick-borne disease of domestic cats in North America. Domestic cats become infected when *C felis* sporozoites are introduced through the bite of infected *Amblyomma americanum*, lone star ticks,[1,2] or *Dermacentor variabilis*, American dog ticks.[3,4] *A americanum* is considered the primary vector due to the corresponding seasonal activity of lone star ticks and occurrence of clinical cases of cytauxzoonosis in domestic cats, overlap in the distribution and abundance of lone star ticks where the majority of *C felis* infections occur in bobcats and domestic cats, and experimental transmission attempts comparing *C felis* transmission between *A americanum* and *D variabilis*.[1,2]

A americanum ticks infected with *C felis* must feed for 36–48 h for transmission of *C felis* sporozoites to

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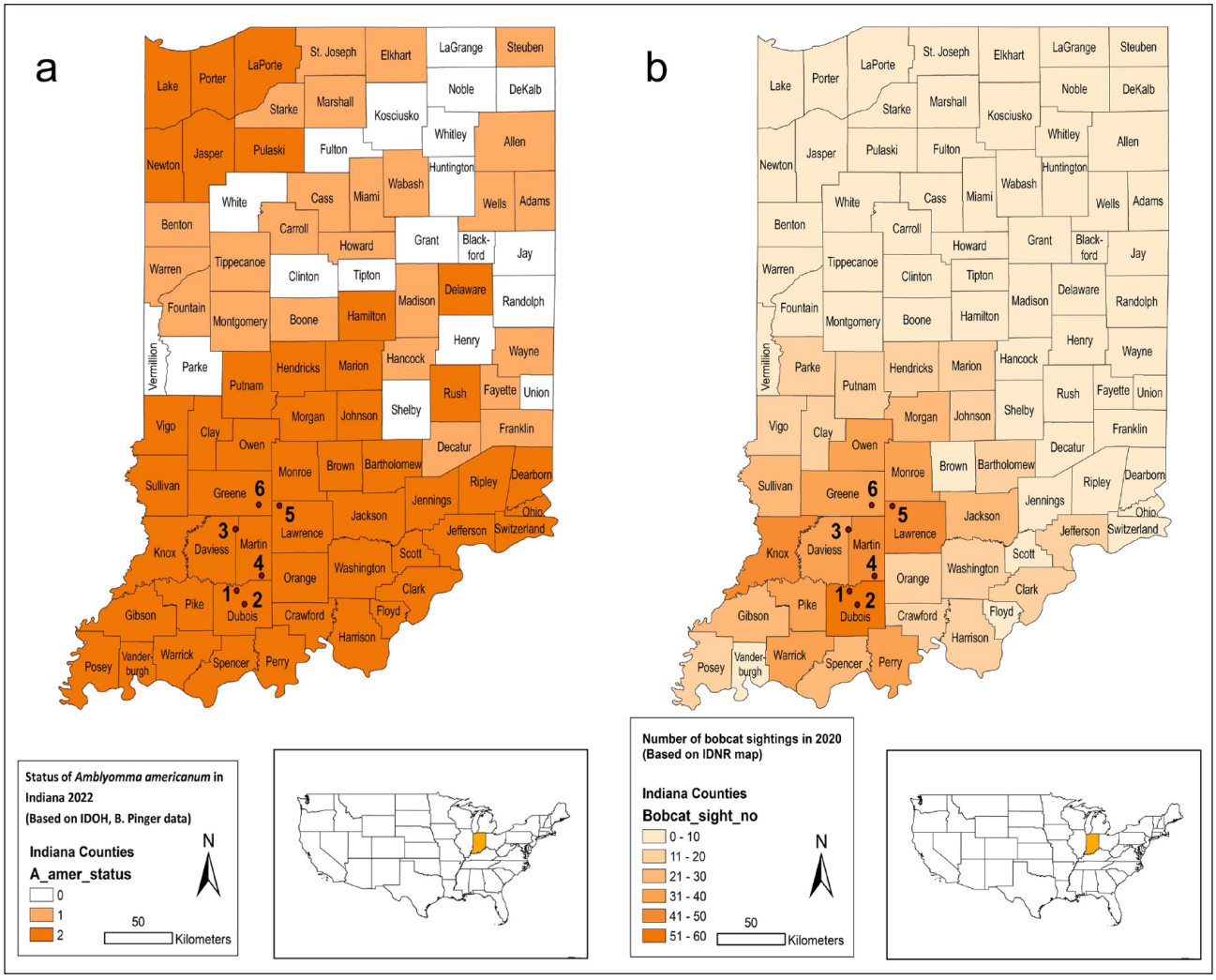

Figure 1 Locations where cases 1–6 of naturally acquired *Cytauxzoon felis* in domestic cats from Indiana occurred in relation to occurrence of (a) *Amblyomma americanum* (lone star ticks) and (b) sightings of *Lynx rufus* (bobcats). In panel (a), for *A. americanum* county status, 0 denotes "not reported," 1 denotes "reported," and 2 denotes "established." Note, *Dermacentor variabilis* is considered established in all Indiana counties

domestic cats.[5,6] Once inside a feline host, sporozoites infect macrophages or other mononuclear cells.[7] Inside host mononuclear cells, *C felis* replicates asexually through schizogony, forming schizonts. Schizonts of *C felis* are considered the most pathogenic phase, and they enlarge and multiply to the point where vessels may become occluded.[8] Clinical signs of acute cytauxzoonosis, including fever, inappetence, lethargy, depression, dehydration, dyspnea, hemolytic crisis and possibly icterus, result from the occlusion of vessels and pro-inflammatory responses.[7] The clinical signs of cytauxzoonosis begin 11–14 days after being bitten by a *C felis*-infected tick.[1,2,5–7]

Since *C felis* was first reported in domestic cats from southwestern Missouri,[9] cases of feline cytauxzoonosis have been documented primarily in the south-central and southeastern USA.[7,10,11] Vertebrate reservoir hosts include chronically infected domestic cats that survived acute cytauxzoonosis, with or without treatment,[12–16] and *C felis*-infected wild bobcats.[17–20] As bobcats are wild animal reservoirs of *C felis* and pyrexia is ubiquitous among cats with acute cytauxzoonosis, the disease is commonly referred to as bobcat fever. The purpose of this retrospective case series is to report cases of cytauxzoonosis from southern Indiana documenting autochthonous transmission of *C felis* to domestic cats in the state for the first time. Veterinary practitioners in Indiana should consider *C felis* infection in cats that are febrile, dehydrated, dyspneic, in hemolytic crisis or icteric.

Case series description

Case 1

Case 1 was a 3-year-old, intact, male domestic shorthair cat that weighed 4.0 kg (8.8 lbs) from Dubois County, Indiana (Figure 1). On 1 June 2018, the cat had 'dark' urine and a rectal temperature of 41.1°C (106.0°F). On the

morning of 2 June 2018, the cat's rectal temperature was 38.9°C (102.0°F), and by evening it was 37.8°C (100.0°F). The cat was euthanized for deteriorating condition. It was submitted to the Heeke Animal Disease Diagnostic Laboratory (HADDL) on 4 June 2018 for a post-mortem examination and histopathology. Grossly, the cat was dehydrated; the mucous membranes and subcutis were icteric; the lungs were congested and edematous with scattered petechiae; the trachea had red-tinged mucus; hemorrhages were present over the epicardium; mesenteric, gastric and mandibular lymph nodes were enlarged and dark red to purple; the urinary bladder contained orange urine; and the spleen was slightly enlarged. Bile and tapeworms, morphologically identified as *Hydatigera* (*Taenia*) *taeniaeformis*, were noted in the stomach (resulting from post-mortem migration from the small intestine). Histopathology revealed schizonts of *C felis* in the lung, spleen, liver, bone marrow, lymph nodes, heart, kidney and brain; thrombi in the spleen and liver; hemorrhage in the lung and lymph nodes; and histiocytosis and erythrophagocytosis in the bone marrow. This cat did not have a known travel history outside the state of Indiana.

Case 2

Case 2 was a 9-year-old, castrated domestic shorthair cat that weighed 5.1 kg (11.2 lbs) from Dubois County, Indiana (Figure 1). The cat presented to the referring DVM (rDVM) on 4 June 2018 for weight loss, weakness and lethargy of several days. A physical examination revealed pyrexia, muffled heart sounds, jaundiced mucous membranes, third eyelid prolapse and a doughy abdomen. A complete blood count (CBC) showed lymphopenia, neutropenia and thrombocytopenia. A chemistry panel showed hypoalbuminemia, hyperbilirubinemia, azotemia, hypocalcemia, hyperglycemia, hyponatremia and hypokalemia. Examination of a Wright–Giemsa-stained thin blood smear revealed merozoites (piroplasms) in erythrocytes (several infected red blood cells per low power field) and few white blood cells. Acute cytauxzoonosis was strongly suspected and treatment with atovaquone-proguanil plus azithromycin was started. The cat died after one dose of therapy. Its rectal temperature at the time of death was 39.1°C (102.4°F). A post-mortem examination was not performed, and tissues were not available post mortem for additional DNA-based testing to confirm or refute *C felis* infection. This cat did not have a known travel history outside the state of Indiana.

Case 3

Case 3 was an 8-year-old, castrated domestic shorthair cat that weighed 6.0 kg (13.2 lbs) from Daviess County, Indiana (Figure 1). The cat presented to the rDVM on 30 May 2019 for anorexia, lethargy and a small subcutaneous abscess on the lateral thorax. Its rectal temperature

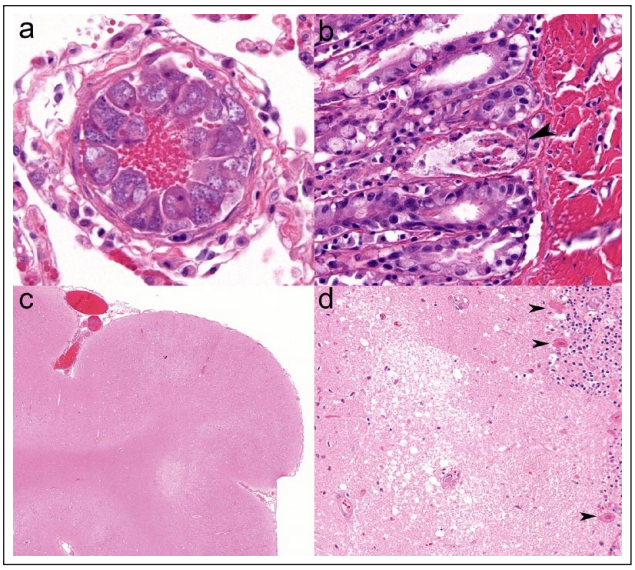

Figure 2 Histopathology of acute cytauxzoonosis, case 3: (a) *Cytauxzoon felis* schizonts within a pulmonary blood vessel. 3. 20× magnification. (b) Intestinal crypt necrosis indicating hypoxia, secondary to microthrombi. 3. 20× magnification. (c) Multifocal areas of cerebral edema. 3. 1.4× magnification. (d) White matter vacuolation indicative of cerebellar edema and Purkinje cell necrosis (arrowheads) also indicative of microthrombi. 3. 15.7× magnification. Slides stained with hematoxylin and eosin

was 39.7°C (103.5°F) and a CBC showed lymphopenia, neutropenia and thrombocytopenia. A chemistry panel showed hyperbilirubinemia. Enrofloxacin therapy was initiated. On 1 June 2019, the cat declined with weakness, marked dehydration and vocalization. Subcutaneous fluid therapy was administered but the cat continued to decline and was euthanized that evening. It was submitted to HADDL on 3 June 2019 for a post-mortem examination and histopathology. Grossly, the cat was dehydrated, the lungs were congested and edematous, and the spleen was congested. Histopathology revealed schizonts of *C felis* in the spleen, liver, kidney, heart, brain and lung (Figure 2). Myocardial necrosis, brain edema and astrogliosis, neuronal necrosis and intestinal crypt necrosis were observed histologically and interpreted as morphologic manifestations of hypoxia and/or microthrombosis. This cat did not have a known travel history outside the state of Indiana.

Case 4

Case 4 was a 2-year-old, spayed domestic shorthair cat that weighed 4.6 kg (10.1 lbs) from Martin County, Indiana (Figure 1). The cat presented to the rDVM on 7 June 2021 for poor balance, ataxia, rapid respiration and anorexia. Before presentation, the cat had been missing for 4 days. On physical examination, the cat was recumbent with nystagmus and tachypnea. A soft tissue swelling was

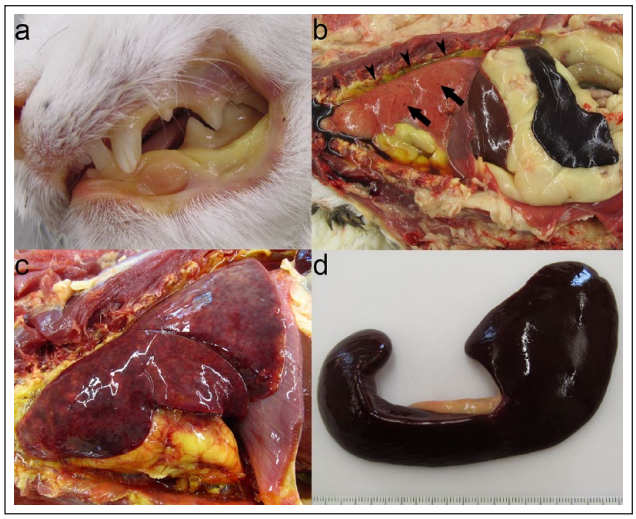

Figure 3 Gross pathology of acute cytauxzoonosis: (a) icterus in gingiva from case 5. (b) Petechiae within the lungs (arrows) and plural effusion (arrow heads) from case 4. (c) Pulmonary edema and congestion from case 5. (d) Splenomegaly from case 5

present over the cat's dorsal thorax. Its rectal temperature was 35.1°C (95.2°F) and a CBC showed lymphopenia, neutropenia and thrombocytopenia. A chemistry panel showed hyperbilirubinemia, azotemia, hypocalcemia, mild hyperphosphatemia, hyponatremia, hypokalemia and mild hypoprotenemia. Thoracic radiographs demonstrated bronchograms. The cat's vaccination history was unknown, and it was SNAP feline immunodeficiency virus/feline leukemia virus negative. The cat was euthanized on 7 June 2021 and submitted to HADDL on 8 June 2021 for a post-mortem examination and histopathology. Grossly, the cat was dehydrated with subcutaneous edema and mild subcutaneous icterus. Pulmonary petechiae, mild pleural effusion, icterus of internal adipose tissue and splenic congestion were also present (Figure 3). Histopathology revealed schizonts in the spleen, lung, liver, kidney, heart, bone marrow and brain, and splenic necrosis and congestion. This cat did not have a known travel history outside the state of Indiana.

Case 5

Case 5 was a 2-year-old, intact male domestic shorthair cat that weighed 4.0 kg (8.8 lbs) from Lawrence County, Indiana (Figure 1). The cat was 'found dead' by the owner. The cat's vaccinations had been current, and it had received a dose of selamectin (Revolution; Zoetis) approximately 1 month previously. The owner indicated that two of their other cats had died some months earlier, in May 2021. The cat was submitted to HADDL on 18 August 2021 for a post-mortem examination and histopathology. Grossly, the gingiva and internal adipose tissue were icteric, the lungs were edematous and congested, and the spleen was congested and enlarged (Figure 3). In addition, the cat was dehydrated, the conjunctiva and subcutis were icteric, the small intestine contained luminal hemorrhage, bilirubinuria was present and the mesenteric lymph nodes were dark red to purple. Histopathology revealed schizonts of *C felis* in the lung, liver, spleen, kidney and brain, as well as splenic erythrophagocytosis, splenic congestion and lymphoid depletion, hepatic cholestasis and pulmonary edema.

Blood samples from five living cats from this same property in Lawrence County were submitted to Oklahoma State University for *C felis* testing on 29 September 2021. Total DNA was extracted from EDTA-anticoagulated blood using the DNeasy Blood and Tissue Kit (QIAGEN) according to the manufacturer's instructions. Extracted DNA was subjected to a *C felis*-specific probe-based droplet digital PCR (ddPCR) using previously described and validated procedures with known positive and negative controls.[21] One of these five cats, a 3.5-year-old, castrated domestic shorthair that weighed 6.8 kg (15.0 lbs), was confirmed to be subclinically infected with *C felis*. The owners reported that they had not observed clinical signs suggestive of cytauxzoonosis in this cat. The cat received a dose of selamectin (Revolution) approximately every month. Neither of the two cats in case 5 had known travel histories outside the state of Indiana.

Case 6

Case 6 was a 3-year-old, intact male domestic shorthair cat that weighed 4.9 kg (10.8 lbs) from Greene County, Indiana (Figure 1). It was an outdoor cat that was allowed to freely roam the neighborhood, and its vaccination history was unknown. The cat presented to the rDVM for weight loss, anorexia of 2–3 days, adipsia, swollen abdomen and lethargy. A physical examination showed the cat was stuporous, blind, hypersalivating, panting, ataxic and infested with ticks. Harsh lung sounds were evident and the mucous membranes were pale. The cat was euthanized by the rDVM and submitted to HADDL on 31 May 2022 for a post-mortem examination and histopathology. Grossly, the cat was dehydrated and anemic, with pulmonary congestion, pleural effusion, splenomegaly, intestinal hemorrhage and lymphadenomegaly/hemorrhage of the abdominal lymph nodes. Tapeworms (unidentified) were present in the small intestine and 4–5 ticks were noted. Histologically, *C felis* schizonts were identified in the lung, liver, sciatic nerve, spleen, lymph nodes, brain, kidney, adrenal gland, heart, intestine and pancreas. In addition, pulmonary edema, hepatic cholestasis and hemorrhage in the sciatic nerve and lymph node were observed histologically.

A subsample of the spleen and ticks from this cat were further evaluated by targeted next-generation sequencing for 21 different vector-borne pathogens following

previously described procedures.[22] Both the spleen and the ticks recovered were confirmed to be infected with *C felis*. The ticks were morphologically identified as *A americanum*. This cat did not have a known travel history outside the state of Indiana.

Discussion

Between June 2018 and May 2022, seven cats from Daviess, Dubois, Greene, Lawrence and Martin counties in southwestern Indiana were diagnosed with *C felis* infection. Populations of *A americanum* are established in most counties in southwestern Indiana as well as a few counties in northwestern Indiana (Figure 1a),[23] whereas all counties in Indiana have established populations of *D variabilis*.[24] Bobcats can occur in almost any county in Indiana but are most common in the southern and west-central regions of the state (Figure 1b).[25] The cases of *C felis* infection reported herein occurred in areas where populations of *A americanum*, *D variabilis* and bobcats are abundant (Figure 1). Bobcats in the central counties in Oklahoma, where tick vectors are known to be common and abundant, were 25.693 times more likely to be infected than in other regions in Oklahoma.[17] Additional studies need to be conducted in Indiana to determine the prevalence and distribution of *C felis* in bobcats, *A americanum* and *D variabilis* to better determine the risk of infection to domestic cats.

Six of the seven cats in the present report had acute cytauxzoonosis. The seventh cat was determined to be a subclinical carrier of *C felis*. Surprising numbers of healthy cats infected with *C felis* have been previously documented in other enzootic states.[12–16,26,27] Future research in Indiana should also determine the prevalence and distribution of subclinical *C felis* infections in domestic cats. It is likely these infected carrier cats serve as domestic reservoirs of *C felis*, posing an additional risk to naive cats.[10]

In the present study, cats infected with *C felis* were domestic shorthairs, with an age range of 2–9 years and 5/6 cats with acute cytauxzoonosis were male. This is consistent with other studies that have reported the majority of cytauxzoonosis cases occur in male cats aged 1–4 years.[12,28] Home ranges of male cats are larger than those of female cats in areas of low-density housing[29] and may represent an increased risk of *C felis* infection. However, cats of either sex or any age are susceptible to infection with *C felis*. Five cases of *C felis* infection were observed in late May and June and two cases were noted in August and September. The occurrence of cytauxzoonosis cases in Indiana was consistent with a seasonal bimodal pattern of occurrence demonstrated in other states[12,29–31] that likely reflects the activity of infected tick vectors feeding on cats. In addition to seasonality, other ecological variables that present increased risks for cats becoming infected with *C felis* include living in low-density residential areas, having access to a wooded habitat, and being near natural or unmanaged areas,[30] all of which provide a habitat for tick vectors and bobcats.

Cats with acute cytauxzoonosis most often present with fever, inappetence, lethargy, depression, dehydration, dyspnea, hemolytic crisis, anorexia and possibly icterus, all of which were observed to varying degrees in the acutely infected cats reported. A diagnosis of cytauxzoonosis is typically based on clinical signs, observation of *C felis* schizonts or merozoites (piroplasm) on stained blood smears, observation of schizonts on histopathology or detection of *C felis* DNA in feline tissues.[7,10] Modern molecular diagnostic methods that detect *C felis* DNA in feline tissues[21,22] are the gold standard and are more sensitive than the observation of *C felis* life stages in tissues. The chronically infected cat in case 5 of the present study was confirmed infected using ddPCR. This method has been validated against other infectious agents that infect cats and specifically detects *C felis* DNA to as low as 0.0000231 ng DNA/reaction in cases of acute cytauxzoonosis and 0.00232 ng DNA/reaction in cases of chronic cytauxzoonosis.[21] ELISA methods that detect *C felis*-specific IgM and IgG, and can differentiate between acute and chronic cytauxzoonosis, have been developed[32] but are not commercially available. A point-of-care test for cytauxzoonosis that could provide accurate, real-time results would be a tremendous asset and allow veterinary practitioners to interpret clinical signs and reach a definitive diagnosis to initiate antiprotozoal therapy promptly.

Recommended therapy for cytauxzoonosis is atovaquone (15 mg/kg PO q8h for 10 days) and azithromycin (10 mg/kg PO q24h for 10 days)[33] along with excellent nursing and supportive care.[7] Cytauxzoonosis was historically considered a highly fatal disease with a poor prognosis. While infection is severe, 60% of cats with cytauxzoonosis treated with atovaquone and azithromycin recover and are 7.2 times more likely to survive to discharge.[33] A prompt diagnosis and the initiation of antiprotozoal therapy early in the course of the disease increases the likelihood of a positive clinical outcome.

Controlling cytauxzoonosis in cats relies on the administration of approved acaricides and limiting the cat's exposure to areas contaminated with ticks. Two products, Seresto (imidacloprid and flumethrin; Elanco)[34] and Revolution Plus (selamectin and sarolaner; Zoetis)[35] have demonstrated efficacy in preventing the transmission of *C felis* to domestic cats from infected ticks. The development of a vaccine that prevents or limits *C felis* infection in cats would provide a critical step for controlling cytauxzoonosis but attempts have met with limited success.[36,37] A vaccination with a replication-defective human adenoviral vector constructed with two *C felis*-specific immunogenic antigens induced cell-mediated and humoral immune responses in cats but did not prevent transmission.[38]

However, immunization significantly delayed the onset of clinical signs and reduced febrility in vaccinated cats exposed to *C felis* infection.

Conclusions

Autochthonous cases of cytauxzoonosis in cats from Indiana were confirmed. Cases of cytauxzoonosis occurred in southwestern Indiana where tick vectors and bobcats are abundant. However, cats in all Indiana counties should be considered at risk as competent tick vectors and bobcats can be found throughout the state. Cats infected with *C felis* had classic signs or clinical abnormalities of cytauxzoonosis, including fever, inappetence, lethargy, depression, dehydration, dyspnea, hemolytic crisis, anorexia, icterus, leukopenia, neutrophilia, thrombocytopenia and hyperbilirubinemia. Common post-mortem findings included dehydration, pulmonary edema, pleural effusion and icterus. The lung, liver, spleen and kidney provided histologic evidence of *C felis* infection in all cases in which tissues were examined histologically. One cat infected with *C felis* was subclinical and was not known to have ever displayed clinical signs of cytauxzoonosis. Treatment for cytauxzoonosis includes administration of atovaquone and azithromycin with excellent nursing and supportive care. Recommendations to control *C felis* infection include preventing access to tick-contaminated habitats and the application of approved acaricides that have demonstrated efficacy for preventing transmission of *C felis* to cats.

Acknowledgements The authors would like to acknowledge the referring veterinarians and clients of the HADDL for providing detailed accounts of the affected cats. Additionally, they thank the technical staff at the HADDL and the Animal Disease Diagnostic Laboratory in West Lafayette for excellent histologic tissue preparation and molecular work.

Author note Some of the findings reported in this publication were presented by GNB at the Purdue Veterinary Conference, 20–24 September 2022, West Lafayette, IN, USA.

Conflict of interest The authors declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding Publication costs were covered through internal institutional mechanisms available to MVR and SD through Oklahoma State University and Purdue University, respectively.

Ethical approval The work described in this manuscript involved the use of non-experimental (owned or unowned) animals. Established internationally recognized high standards ('best practice') of veterinary clinical care for the individual patient were always followed and/or this work involved the use of cadavers. Ethical approval from a committee was therefore not specifically required for publication in *JFMS*. Although not required, where ethical approval was still obtained it is stated in the manuscript.

Informed consent Informed consent (verbal or written) was obtained from the owner or legal custodian of all animal(s) described in this work (experimental or non-experimental animals, including cadavers) for all procedure(s) undertaken (prospective or retrospective studies). No animals or people are identifiable within this publication, and therefore additional informed consent for publication was not required.

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