



Eosinophilic pericardial effusion and pericarditis in a cat

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Abstract

Case summary A 10-year-old domestic shorthair cat presented for lethargy, anorexia and labored breathing. Significant pleural and pericardial effusions prompted thoracocentesis and pericardiocentesis. Cytologic evaluation of the pericardial effusion revealed a highly cellular hemorrhagic, eosinophilic (12%) effusion, with many markedly atypical suspected mesothelial cells, interpreted as concerning for neoplasia. Thoracoscopic subtotal pericardiectomy and histology of the pericardium revealed predominantly eosinophilic inflammation with multifocal mesothelial hypertrophy and ulceration. A peripheral eosinophilia was not present on serial complete blood counts. Initial infectious disease testing was mostly negative. *Toxoplasma gondii* titers were most consistent with prior exposure, although reactivation could not be excluded. The owner's medical history included a prior diagnosis of bartonellosis. Owing to the challenges of definitive *Bartonella* species exclusion, the cat was treated empirically with pradofloxacin and doxycycline, and a subtotal pericardiectomy. There was improvement at first but pleural effusion recurred approximately 3 months after discharge. The cat was euthanized and a necropsy was not performed. Subsequent pericardial effusion *Piroplasma/Bartonella/Borrelia* droplet digital PCR detected DNA of *Bartonella vinsonii* subspecies *berkhoffii*, and peripheral blood culture and sequencing revealed a rare apicomplexan organism (90% homology with *Colpodella* species) of unknown clinical significance. Testing for filamentous bacteria and fungal pathogens was not performed.

Relevance and novel information This case offers several unique entities – eosinophilic pericardial effusion and eosinophilic pericarditis of unknown etiology – and illustrates the well-known marked atypia that may occur in reactive and hyperplastic mesothelial cells, particularly of infrequently sampled and cytologically described feline pericardial effusion, supporting a cautious interpretation of this cytology finding.

Keywords: Apicomplexan, *Bartonella*; eosinophilic effusion; mesothelial cells; pericardial effusion; pericarditis

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Case description

A 10-year-old female spayed domestic shorthair cat was referred to the North Carolina State University Veterinary Hospital (NCSU-VH) for evaluation of a 4-day history of lethargy, decreased appetite and labored breathing. The cat lived indoors with minimal outdoor access, intermittently received topical selamectin (Revolution; Zoetis) and was current on vaccinations. Two owners had tested positive for COVID-19, 6 weeks before the cat's presentation.

Physical examination revealed increased respiratory effort and muffled heart sounds. Owing to the owner's

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Table 1 Selected effusion analysis results

| Test | Pleural | Pericardial |
|-----------------------------------|---------|-------------|
| HCT (%) | 0.2 | 10.9 |
| NCC ($\times 10^3/\mu\text{l}$) | 0.40 | 29.05 |
| TP (g/dl) | <2.5 | 3.5 |
| 100-cell leukocyte differential | | |
| Neutrophils (%) | 51 | 69 |
| Macrophages (%) | 25 | 6 |
| Lymphocytes (%) | 22 | 13 |
| Eosinophils (%) | 2 | 12 |

HCT = hematocrit; NCC = nucleated cell count (ADVIA 120, Siemens); TP = total protein (refractometer)

history of COVID-19 infection, per hospital policy, the cat was hospitalized in isolation, limiting initial diagnostics pending a severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) PCR test. An echocardiogram performed by a cardiologist identified 1.5cm of hypoechoic pericardial effusion, and a small quantity of pleural effusion (140ml of pleural effusion was removed before referral). The cat was hemodynamically stable and there were no cardiac structural abnormalities. Overnight the cat clinically worsened, with tachypnea progressing to dyspnea, and was hypotensive. Point-of-care ultrasound (POCUS) was concerning for tamponade, prompting therapeutic pericardiocentesis. The patient was administered intravenous (IV) butorphanol, ketamine and alfaxalone for sedation and analgesia. Approximately 60ml of serosanguineous pericardial effusion and a small quantity of pleural effusion were obtained and submitted for evaluation.

The pericardial effusion was markedly cellular, with a nucleated cell count and total protein consistent with a hemorrhagic exudate with increased eosinophils (Table 1). Abundant individualized and aggregated large round atypical cells were noted with moderate to marked anisocytosis (Figure 1). Frequent multinucleated cells (up to nine nuclei) displaying moderate to marked anisokaryosis, with a large prominent nucleolus, and occasional nuclear molding were seen (Figures 2 and 3). Cells had large amounts of deep blue cytoplasm, with low numbers displaying a pink cytoplasmic fringe. The cytologic interpretation was eosinophilic hemorrhagic effusion with atypical cell population concerning for neoplasia, although the possibility of a highly dysplastic mesothelial population secondary to inflammation or fluid chronicity was not excluded.

The pleural effusion was interpreted as a transudate (Table 1), with low numbers of suspected reactive mesothelial cells, although a neoplastic population could not be excluded. Pleural effusion aerobic and anaerobic bacterial culture yielded no growth. Fungal culture was not performed.

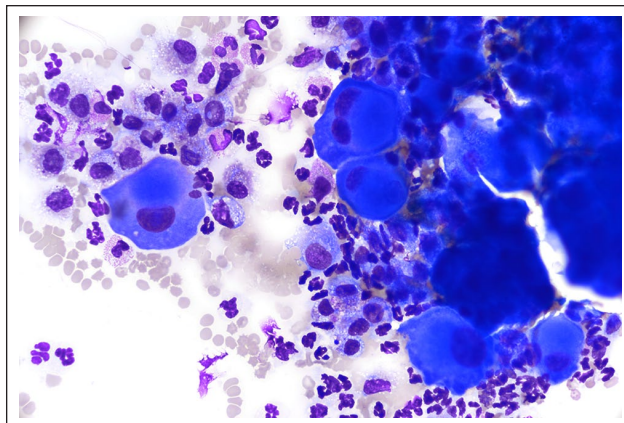


Figure 1 Pericardial effusion, direct smear, Wright Giemsa stain, $\times 50$. Individualized and dense aggregates of atypical cells with large nuclei with stippled chromatin, prominent nucleoli and large amounts of medium to deep blue cytoplasm. Vacuolated macrophages, neutrophils and increased numbers of eosinophils present

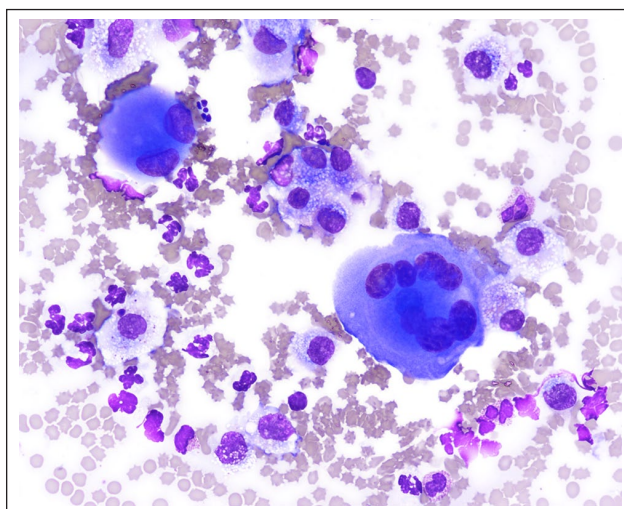


Figure 2 Pericardial effusion, concentrated direct smear, Wright Giemsa stain, $\times 50$. Large atypical cells displaying bi- and multinucleation with moderate anisokaryosis and prominent nucleoli. Vacuolated macrophages, variably disrupted neutrophils and eosinophils present

A negative SARS-CoV-2 PCR (nasal swab sample) result received on the third day of hospitalization permitted further diagnostic imaging. Radiographic findings included right cranial lung lobe collapse and caudoventral mediastinal opacity. Abdominal ultrasound revealed no significant abnormalities. Thoracic CT revealed persistent moderate pericardial effusion with mild pericardial thickening and a small volume of pleural effusion. The patient had remained clinically stable after the initial thoracocentesis and pericardiocentesis, but the persistent pericardial effusion prompted recommendation of diagnostic and therapeutic

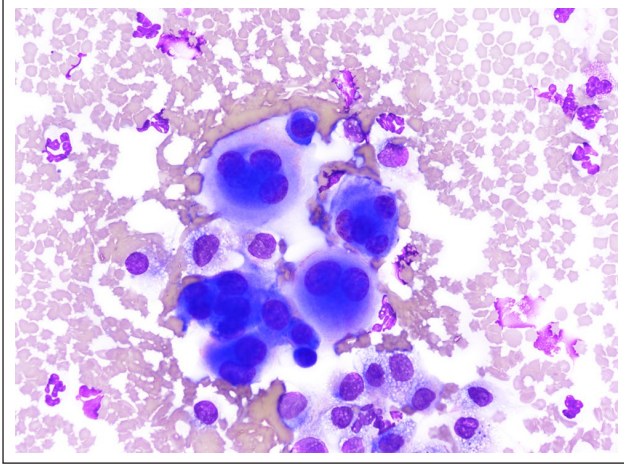


Figure 3 Pericardial effusion, concentrated direct smear, Wright Giemsa stain, $\times 50$. Atypical multinucleated cells displaying mild anisokaryosis, prominent nucleoli, nuclear molding and faint pink cytoplasmic fringe

pericardiectomy, as well as *Bartonella* species serology (NCSU-VH). The owner reported that several years prior, she was diagnosed with bartonellosis after a cat scratch from the patient or a feline housemate. After a right lateral thoracoscopic subtotal pericardiectomy was performed with an uneventful recovery, the patient received a surgical site injection of bupivacaine liposome suspension (Nocita; Elanco) and was maintained on constant rate infusions of IV fentanyl and ketamine. An 8-week treatment course for *Bartonella* species (pradofloxacin, doxycycline) was initiated. Throughout hospitalization, serial complete blood counts (ADVIA 120) were performed and a peripheral eosinophilia was never documented. There were no significant hematologic, biochemistry or urinalysis findings. The cat was discharged 9 days after the original presentation.

Surgical histopathology revealed marked expansion of the parietal pericardium by granulation tissue (Figure 4), with robust perivascular infiltrates of inflammatory cells, primarily eosinophils, overlain by plump mesothelial cells (Figures 5 and 6). The morphologic diagnosis was marked, chronic, diffuse eosinophilic, neutrophilic and lymphoplasmacytic fibrinous pericarditis with multifocal mesothelial hypertrophy, ulceration and chronic hemorrhage. There was no evidence of neoplasia or infectious agents. Immunohistochemical staining of the pericardium for feline coronavirus (feline infectious peritonitis [FIP]), Warthin-Starry for argyrophilic infectious organisms, and Gomori methenamine silver (GMS) and Periodic acid–Schiff (PAS) special staining for protozoal and fungal organisms were negative.

The original infectious disease test results are summarized in Table 2. In the absence of an alternative diagnosis and the difficulty in definitive exclusion of *Bartonella* species infection, the patient completed an 8-week course of pradofloxacin and doxycycline per the

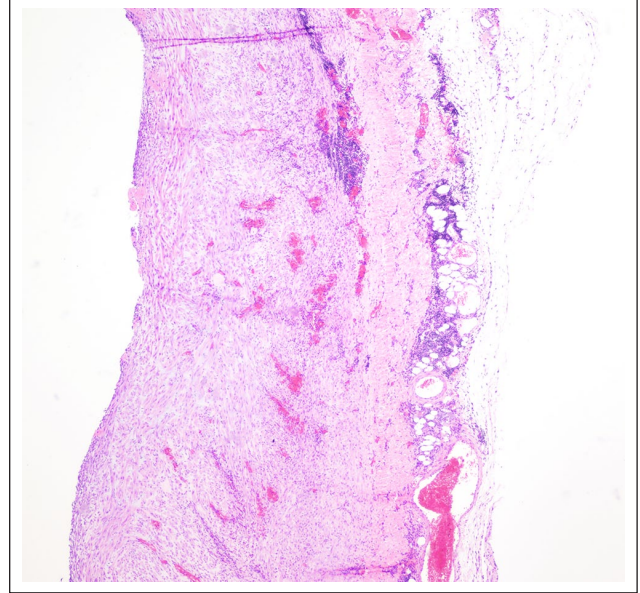


Figure 4 Parietal pericardium, hematoxylin and eosin, $\times 4$. Parietal surface (left) is markedly expanded by granulation tissue with small perpendicularly oriented vessels

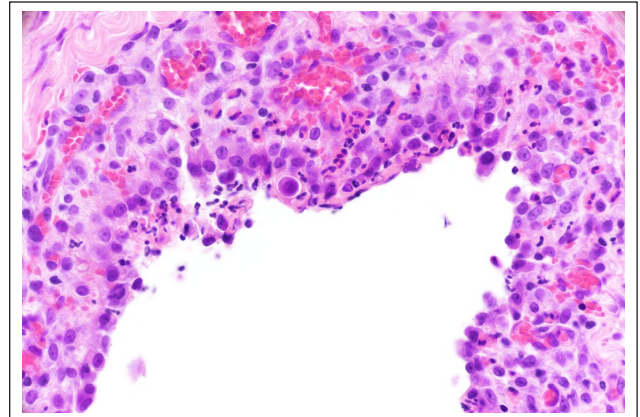


Figure 5 Parietal pericardium, hematoxylin and eosin, $\times 50$. Visceral surface of the parietal pericardium displaying focal hypertrophy of plump mesothelial cells with infiltrating eosinophils

recommendation of the Vector Borne Disease Lab at NCSU-VH. Paired *Toxoplasma gondii* titers revealed a persistently weakly positive IgM titer, and the lack of increase in the IgG titer was interpreted as most consistent with prior exposure or potential *Toxoplasma* reactivation (Table 3). The cat initially improved, with no evidence of recurrent effusion on thoracic POCUS on recheck examination at NCSU-VH 4 weeks after pericardiectomy. However, 3 months after discharge, recurrent pleural effusion was noted at another hospital, which was managed via thoracocentesis. Soon after, the patient was euthanized; a necropsy was not performed.

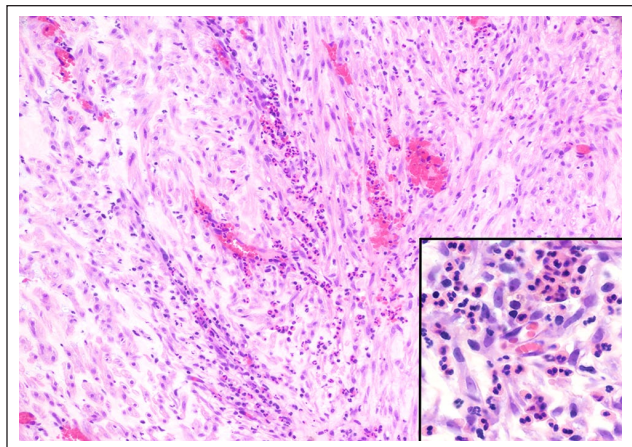


Figure 6 Parietal pericardium, H&E, ×20. Abundant immature fibroblasts admixed with small caliber blood vessels and predominantly eosinophilic infiltrate. Inset: Pericardium, H&E, ×50. Fibroblasts with marked eosinophilic infiltrate, centrally small caliber vessels. H&E = hematoxylin and eosin

Table 2 Selected original infectious disease testing results

| Test | Result |
|---|---|
| FelV – antigen (IDEXX Feline Triple SNAP) | Negative |
| FIV – antibody (IDEXX Feline Triple SNAP) | Negative |
| <i>Dirofilaria immitis</i> – antigen (IDEXX Feline Triple SNAP) | Negative |
| <i>Dirofilaria immitis</i> – antibody (IDEXX ELISA) | Negative |
| SARS-CoV-2 (PCR, NCSU) | Negative |
| Influenza A (PCR, NCSU) | Negative |
| <i>Bartonella</i> species serology (IFA, NCSU) | Negative <i>B. vinsonii</i> <1:32 <i>B. henselae</i> <1:32 <i>B. koehlerae</i> <1:32 |
| <i>Hepatozoon</i> species (IDEXX RealPCR) | Negative |
| <i>Borrelia burgdorferi</i> (IDEXX PCR) | Negative |
| <i>Anaplasma phagocytophilum</i> (IDEXX PCR) | Negative |

FelV = feline leukemia virus; FIV = feline immunodeficiency virus; IFA = immunofluorescent antibody

Research results using a previously validated *Piroplasma/Bartonella/Borrelia* droplet digital PCR assay (ddPCR)¹ on pleural effusion, pericardial effusion and pericardium returned postmortem, due to COVID-related delays in obtaining reagents. This detected

Table 3 Serum titer results for *Toxoplasma gondii*

| <i>Toxoplasma gondii</i> | Original* IFA-IDEXX | 22 days later† ELISA-CSU |
|--------------------------|------------------------|-----------------------------|
| IgG | Positive 1:1600 | Positive 1:1024 |
| IgM | Positive 1:50 | Positive 1:64 |

*Initial titer submitted by the primary care veterinarian shortly after discharge from NCSU

†Recheck titer submitted by NCSU-VH to CSU

CSU = Colorado State University; IFA = immunofluorescent antibody; NCSU-VH = North Carolina State Veterinary Hospital

DNA of *Bartonella vinsonii* subspecies *berkhoffii* in the pericardial effusion. *Piroplasma* DNA was amplified from a 14-day liquid (Brugge Medium)² enrichment culture of the peripheral blood. After DNA concentration and qPCR testing, the partial 18S rRNA gene DNA sequence had 90% homology with an uncultured apicomplexan, *Colpodella* (GenBank accession FJ410758).

Discussion

Pericardial effusion is infrequently encountered in cats, with necropsy reports indicating an incidence of 1–2.3%.^{3,4} Small volumes are often noted in cats with congestive heart failure secondary to underlying cardiac disease (various cardiomyopathies), the most common cause of feline pericardial effusion.^{5,6} Neoplastic causes include lymphoma, carcinoma and mesothelioma. Infectious causes include FIP, bacterial pericarditis, cryptococcosis, toxoplasmosis, feline leukemia virus and panleukopenia. Inflammatory diseases, uremia and coagulopathies have also been linked to pericardial effusion.^{3–6} Significant volumes with the potential to cause cardiac tamponade are very rare in cats.⁶ While they may occur due to underlying cardiac disease, large volumes are usually associated with neoplastic or infectious causes.^{5,7,8} In our patient, the development of tamponade was suspected given the large volume of pericardial effusion observed on thoracic POCUS before centeses. It is possible that recurrent pleural effusion contributed to the clinical decompensation.

Feline pericardial effusions are infrequently sampled due to characteristically small volumes and frequent association with cardiomyopathy. When volumes or clinical signs prompt therapeutic or diagnostic pericardiocentesis, reported cytologic findings are limited to the most relevant diagnostic features (eg, lymphoblasts, infectious agents) or brief interpretive findings (eg, chylous effusion, mesothelial hyperplasia).^{5,7,8} The primary utility of pericardial effusion cytology is considered evaluation for lymphoma, inflammatory or infectious disease. Exfoliated mesothelial cells are an expected

finding, with reactive or dysplastic cells often displaying criteria of malignancy, rendering definitive differentiation between reactive mesothelial cells and neoplastic cells (chiefly carcinoma or mesothelioma) difficult to impossible in many cases.^{9,10} In this case, dense clusters of more typical reactive-appearing mesothelial cells were admixed with frequent large, atypical cells. Criteria of malignancy observed included multinucleation, marked anisocytosis, marked anisokaryosis, nuclear molding and crowding, prominent nucleoli and anisonucleoliosis. These findings, coupled with the clinical presentation, prompted an interpretation of atypical cells concerning for neoplasia, with primary differentials of mesothelioma and carcinoma.

Histopathologic evaluation of a sufficiently large tissue sample is necessary to differentiate reactive mesothelial hyperplasia from neoplasia, with immunohistochemistry often used in further characterization of a neoplastic population.¹¹ Thoracoscopic subtotal pericardiectomy was pursued in this case for therapeutic and diagnostic purposes. Two relatively large tissue sections (up to $2.5 \times 2.5 \times 0.2$ cm) were submitted, allowing for confident assessment of the pericardial changes and exclusion of a neoplastic process affecting the pericardium. The marked atypia of the mesothelial cells on cytology was absent in the histologic sample obtained approximately 6 days later.

Eosinophilic pericarditis has not, to our knowledge, previously been reported in a cat. This finding was consistent with the eosinophilic (12%) character of the pericardial effusion. Eosinophilic cavity effusions ($\geq 10\%$ eosinophils) are uncommon in animals, and are often associated with neoplastic conditions.¹²⁻¹⁴ A recent case report describes what the authors believe to be the first published eosinophilic pericardial effusion in a cat, which ultimately was suspected to be a paraneoplastic response with concurrent eosinophilia due to probable lymphoma.¹⁵ Reported infectious causes of eosinophilic effusions include heartworm disease, fungal infection and lung worms.^{9,13,16,17} A fungal etiology was considered less likely given the patient's initial clinical improvement after empiric antibiotic therapy and pericardiectomy. Further, the use of special stains (GMS and PAS) on pericardium tissue sections would be expected to increase histologic detection of any organisms present; however, additional more sensitive diagnostics to further evaluate this possibility (eg, fungal culture and/or panfungal PCR of effusions) were not pursued. Fecal flotation and a tracheal wash could have been performed to screen for endoparasites, and hypersensitivity or parasitic airway diseases, respectively. These were considered less likely (although cannot be

excluded) given the cat's clinical presentation, history and initial improvement in the absence of anti-inflammatory/immunomodulatory or anthelmintic treatment. Eosinophilic inflammatory tissue infiltrates may be seen in patients with hypereosinophilic syndrome or eosinophilic leukemia.¹⁸ Neither fit the clinical picture in this cat, given the absence of a peripheral eosinophilia, which is typically marked in either disease process.

Cats are the natural reservoir host for several *Bartonella* species, found in both healthy cats and those with associated lesions, including endocarditis and endomyocarditis.¹⁹⁻²¹ Frequently utilized diagnostics include serology, culture (via *Bartonella* alpha Proteobacteria growth medium), PCR and tissue immunohistochemistry; however, confident exclusion of *Bartonella* species infection remains elusive.^{1,20}

In recent years, Chinese investigators have reported infection with members of the phylum Apicomplexa that are most closely related to *Colpodella* species in *Dermacentor* ticks, a tiger, horses and two human patients – one with a chronic history of malaise, hemolytic anemia and intra-erythrocytic organisms, and the other with neurological symptoms.²² Phylogenetically, *Colpodella* species are closely related to the pathogenic protozoa *Sarcocystis*, *Toxoplasma* and *Babesia* species.²³ Whether the sequence obtained from the enrichment blood culture from this cat is a *Colpodella* species and/or is of clinical relevance in the context of the eosinophilic pericarditis and effusion in this cat is unknown.

Owing to the pending ddPCR results, occasional patient outdoor access, owner history of bartonellosis and the inherent challenges of bartonellosis exclusion, an 8-week course of pradofloxacin and doxycycline was completed. It is unclear if the patient's initial improvement was due to the response to antibiotic treatment of *Bartonella* species (or another undetected bacterial etiology) or related to the therapeutic effect of the pericardiectomy. In the light of the ddPCR results and recurrence of the pleural effusion, an infectious etiology remains a consideration.

Conclusions

Recurrence of the pleural effusion may have been due to an unknown infectious etiology, unidentified neoplasia or an unidentified inflammatory condition. Lacking a post-mortem examination, the etiology of the eosinophilic pericarditis and effusion remains unknown. The marked atypia of the reactive mesothelial cells on cytology and their absence in a histologic sample offers a cautionary example of the challenges inherent to pericardial effusion cytology interpretation, particularly in infrequently described feline samples.

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Ethical approval The work described in this manuscript involved the use of non-experimental (owned or unowned) animals. Established internationally recognised 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 Open Reports*. 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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