Granulomatous pneumonia caused by *Mycobacterium simiae complex* and necroulcerative enteritis caused by *Entamoeba* spp in a panther chameleon (*Furcifer pardalis*)

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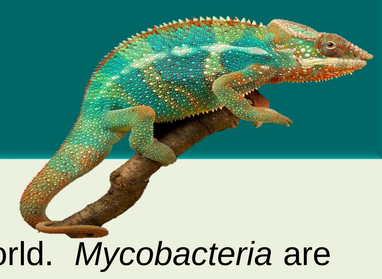


INTRODUCTION

The panther chameleon (*Furcifer pardalis*) is a species of Chamaleonidae native to Madagascar, commonly kept as a pet in many parts of the world. *Mycobacteria* are ubiquitous in nature and can be isolated from soil, water and bioaerosols. Mycobacteriosis is considered a common condition in captive reptiles and it is usally associated with immunosupression. Common species affecting reptiles include *Mycobacterium marinum*, *M. fortuitum* and *M. chelonae*. Amebiasis has also been commonly reported as a cause of entercholitis in reptiles, common species are *Entamoeba invadens* and *E. histolytica*. Reptiles generally acquire mycobacteria and amoebae via ingestion. A 9-month-old male captive Panther chameleon (*Furcifer pardalis*) was presented with history of dehydration, dyspnea, anorexia, and diarrhea.

MATERIAL AND METHODS

This case report describes the pathological aspects observed in this chameleon and the molecular identification of the aetiological agent. A complete necropsy of the animal was performed and samples for histopathology were taken. Due to the evidence of an infectious process, histochemical stains, including PAS, Gram, and Ziehl-Neelsen were also applied. Additionally, PCR for *Mycobacterirum* spp using total DNA extracted from formalin-fixed paraffin-embedded (FFPE) lung tissues and sequencing were performed.

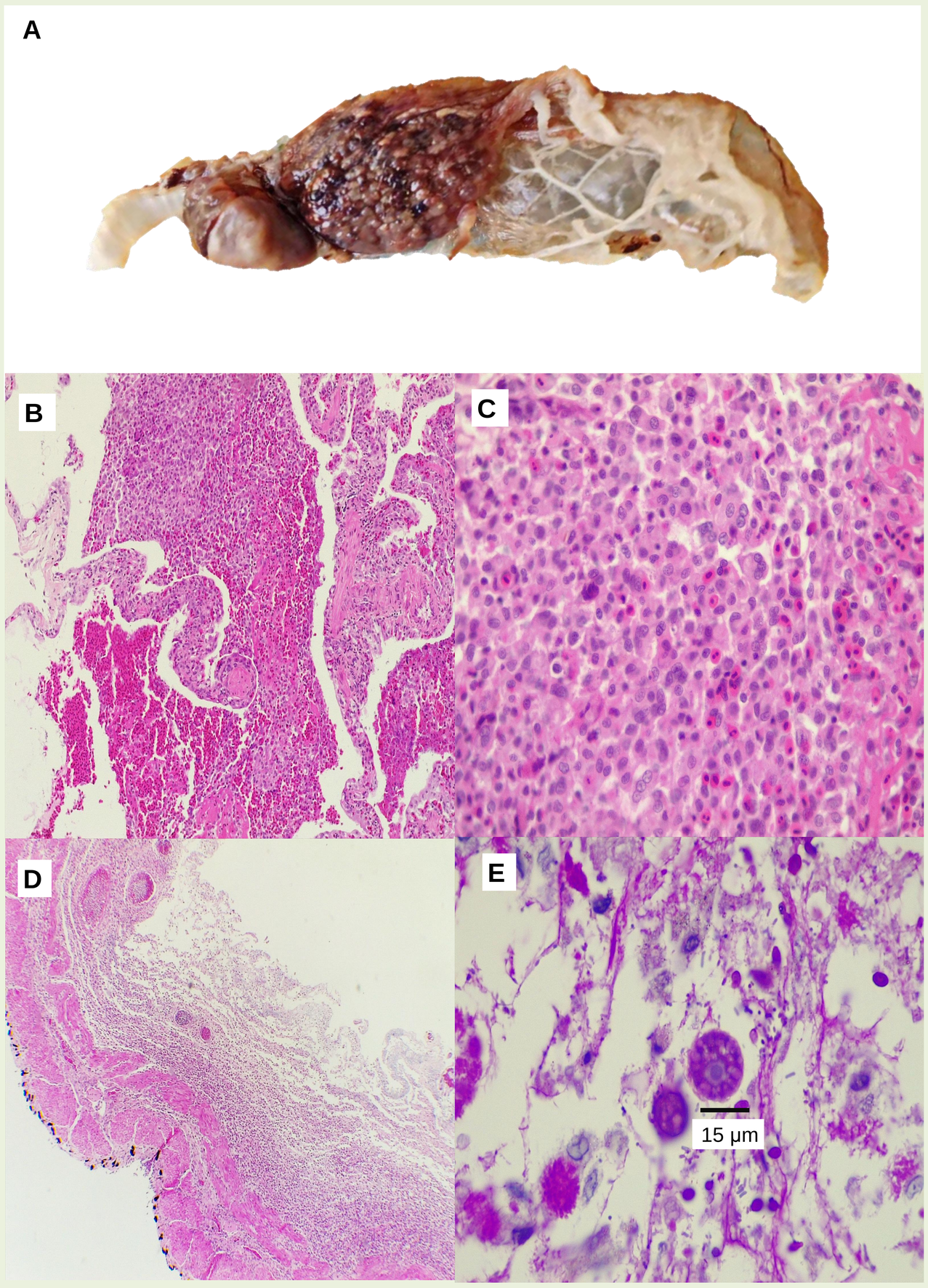
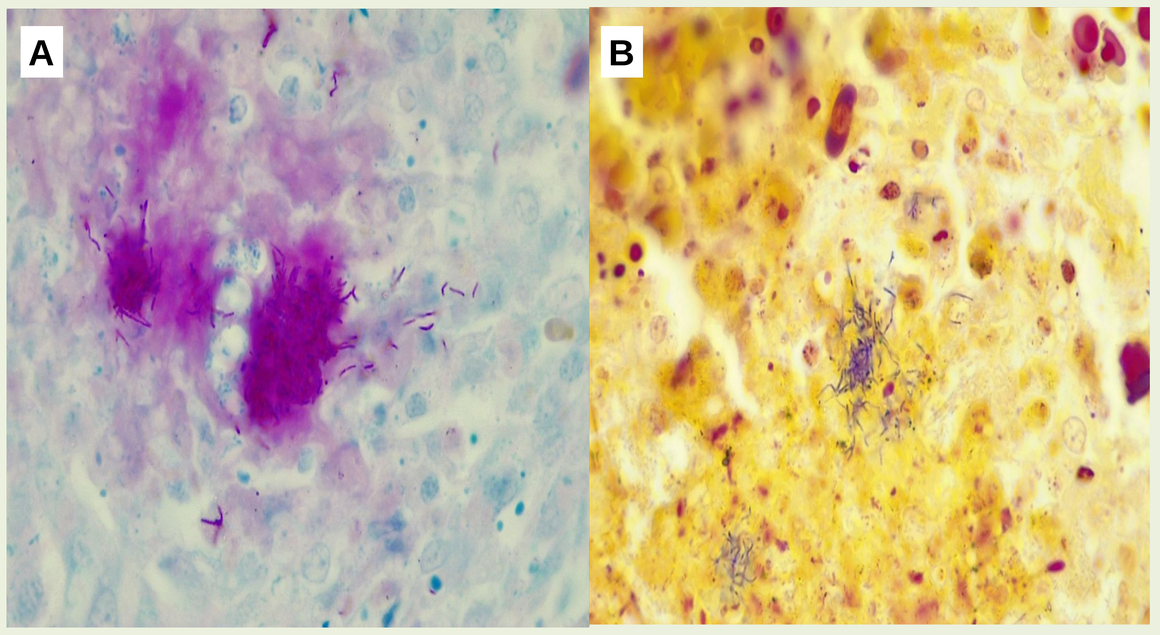


Figure 1. A. Gross appareance of the lungs. The surface presents numerous coalescent nodules.
B. Photomicrography of the lung. H&E, 4x. The lumen of faveolar septa are expanded by necrotic debris, aggregates of epithelioid macrophages and hemorrhage. **C.** Photomicrography of the lung. H&E, 40x. Numerous epitheloid macrophages and multinucleated giant cells in the lumen of the faveolar spaces.

D. Photomicrography ot intestine. H&E,4x. Necroulcerative enteritis. The lumen of the intestine is filled with necrotic debris. Copious amounts of macrophages and heterophils infiltrate the submucosa and extend to the muscular layer. **B.** Photomicrography of intestine. PAS, 100x. Presence of ameboic throphozoites admixed with bacteria. The PAS positive trophozoites average 10-16 micrometers in diameter

RESULTS

Macroscopically, the lungs presented multiple coalescent greyish-white nodules, and the intestine was severely distended. Histopathology revealed a severe and generalized necrohaemorrhagic and granulomatous bronchopneumonia with Gram-positive bacteria and intralesional acid-fast bacilli; the intestine exhibited a necrotic and ulcerative enterocolitis with evidence of intralesional 15-20 microns diameter, PAS positive amoebic trophozoites. PCR confirmed the presence of *Mycobacterium* spp by amplifying the mycobacterial 16S rRNA gene. Sequencing of the amplified fragment identified *Mycobacterium simiae complex*.



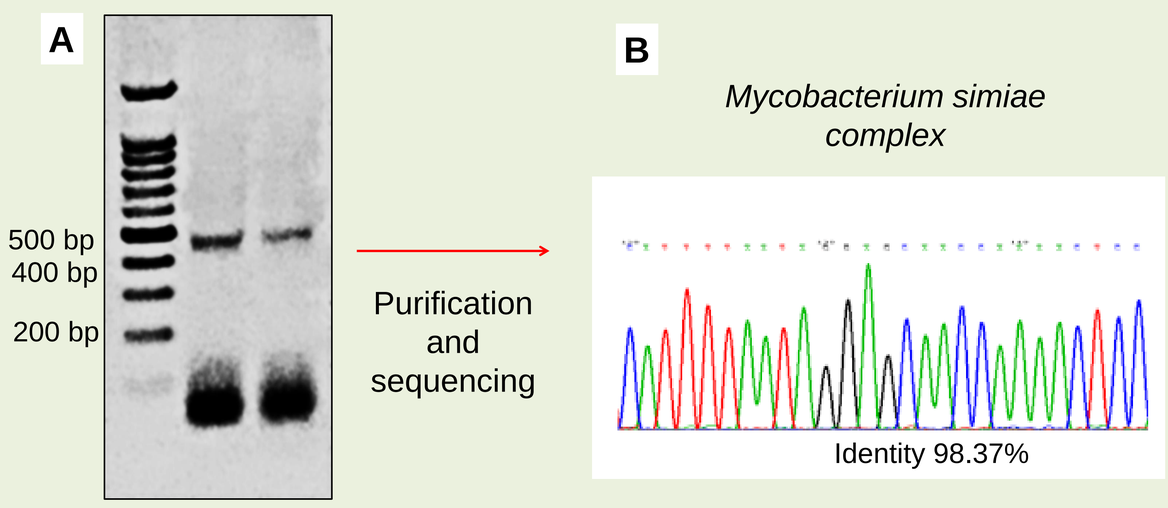


Figure 2. Photomicrography of the lung **A**. Intralesional acid-fast bacilli. Ziehl Neelsen 100x. **B.** Intralesional Gram positive bacteria. Gram, 100x.
Figure 3. Identification of the aetiological agent**. A.** A 500 bp fragment of the 16s ribosomal gene of mycobacteria was amplified. **B.** Sequence analysis revealed that the fragment has 98.37% identity with bacteria of the *Mycobacterium simiae complex.*

CONCLUSIONS

This case shows a polymicrobial infection in a chameleon; the granulomatous bronchopneumonia and the ulcerative enterocolitis are consistent findings in infections by mycobacteria and entamoeba. Non-tuberculous mycobacterias including *M. avium, M. chelonae, M. fortuitum, M. marinum, M. haemophilum, M. kansasii*, and *M. ulcerans* have been reported to be etiologic agents that cause histiocytic granulomas in reptiles and should top the list of differential diagnoses for lesions similar to this case. Amebas are common commensal parasite of different free-ranging reptiles and usually are associated with inmunosupresion. To our knowledge, this is the first report of granulomatous bronchopneumonia in a reptile caused by *Mycobacterium simiae complex.*

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