

## CASE REPORT

# GEOTRICHOSIS CONCURRENT WITH BACTERIAL INFECTION IN A CAPTIVE ESTUARINE CROCODILE

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**ABSTRACT.** A male estuarine crocodile (*Crocodylus porosus*) from a private farm was necropsied. The findings revealed severe suppurative necrotizing pneumonia and pericarditis. The thymus, heart, right lung, liver, and cloacal swab were subjected to microbiology examination. *Geotrichum candidum* and *Pseudomonas aeruginosa* were isolated from the right lung, heart, and liver. The thymus and cloacal swab were positive for *Escherichia coli*. The former was also positive for *Klebsiella pneumoniae*. The cause of death was most likely due to circulatory and respiratory failure that resulted from geotrichosis with concurrent mixed bacterial infection. This paper describes the laboratory findings and possible route of infection that leads to the infection. This organism has a zoonotic potential especially in immunocompromised individuals particularly those that had close contact with the infected animal.

**Keywords:** *Geotrichum candidum*, zoonotic, geotrichosis, fungal pneumonia, crocodile

## INTRODUCTION

The estuarine crocodile (*Crocodylus porosus*) inhabits coastal brackish water, swamps, inland lakes, and marshes of the Indo-Pacific which include Malaysia (van Dijk, 1998). It is the largest size amongst crocodylian species and known by Malaysians as *buaya tembaga* or *buaya katak*. It is listed in Appendix II in the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). Thus, the trade of this species is controlled. There are seven estuarine crocodile farms that are registered with CITES in Malaysia, which are located in Johor (1), Kedah (1) Sabah (3) and Sarawak (2). These farms operate primarily to produce leather and as tourism attraction sites. In October 2019, an adult estuarine crocodile from a farm was presented to Veterinary Laboratory of Southern Zone (MVZS) for necropsy.

*Geotrichum candidum* is commonly isolated in soil, plant substrates, water, and milk (Ellis,

2001) and present worldwide. This fungus is well known to the French cheesemakers as it is used in production of wash-rinds and bloomy rind cheeses (Marcellino *et al.*, 2001). Nevertheless, this organism has been reported to cause invasive skin infection in human (Sfakianakis *et al.*, 2007), fungal septicemia in immunocompromised human (Ng *et al.*, 1994), tonsillitis in pig (Lee *et al.*, 2011), enteritis in dog (Lee *et al.*, 2010) and abortion in cattle (Antoniassi *et al.*, 2013). The current paper reports a case of an estuarine crocodile in captivity that had died due to concurrent geotrichosis and mixed bacterial infection.

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The animal was kept with another 84 crocodiles. According to the submitter, it had a history of intermittent inappetence for two years. This male estuarine crocodile was found dead and sent for necropsy in Veterinary Laboratory of Southern Zone (MVZS).

## DIAGNOSTIC WORKOUT

The heart, right lung, thymus, liver and cloacal swab were cultured on blood and MacConkey agar. Identification of bacteria includes observation of the colony morphology, Gram staining and series of biochemical tests. The samples were also cultured on Sabouraud Dextrose Agar. Later, the fungal colony was transferred into sterile water and sent to the Veterinary Laboratory Services Unit, Faculty of Veterinary Medicine, Universiti Putra Malaysia for species identification.

## FINDINGS

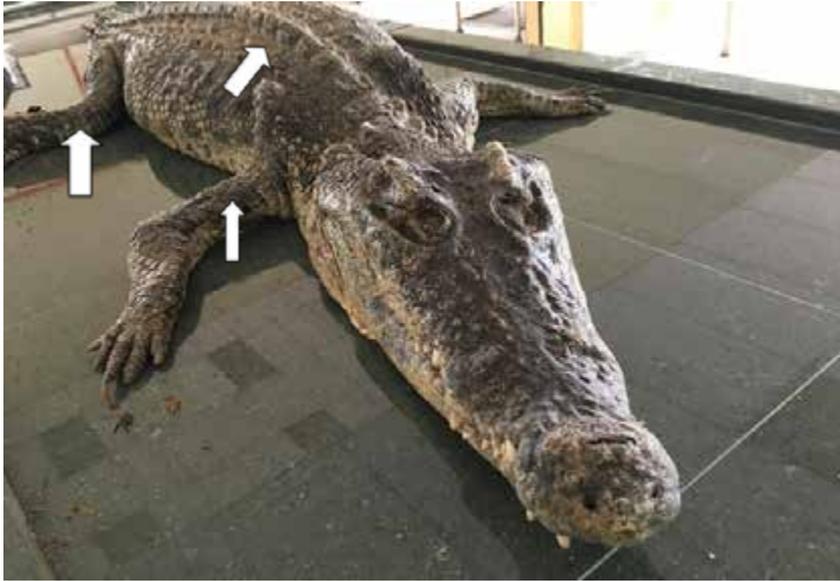
The animal was three meters long and cachectic as evidenced by Figures 1a to 1c. A 15 cm maggot infested-wound at the left quadrato/articular joint was noticed in (Figure 2). In addition, two puncture wounds about 2 to 3 cm in diameter were noted 3 cm cranial to the cloaca (Figure 3a to 3b). The left pleural cavity was empty, and the left lung had turned into oily, black mushy tissue. The left thoracic wall was infested by whitish velvety fungus (Figure 4a to 4b). Meanwhile, the pleura of the right lung had numerous whitish nodules ranging from 0.2 to 2 cm in diameter (Figure 5). The thymus was noticed as a whitish lobulated organ at the base of the heart. The pericardial sac had thickened and presence of numerous whitish



**Figure 1a.** The carcass had a thin neck and shrunken jowls.

to yellowish pus were noted on the epicardium (Figure 6a to 6c). The liver, spleen and kidney were soft and blackish, most likely due to post-mortem changes. These findings were expected as the animal was dead approximately 12 hours

prior to submission for necropsy. *Geotrichum candidum* and *Pseudomonas aeruginosa* were isolated from the right lung, heart and liver. *Geotrichum candidum* showed a good growth at room temperature as white, fluffy colonies



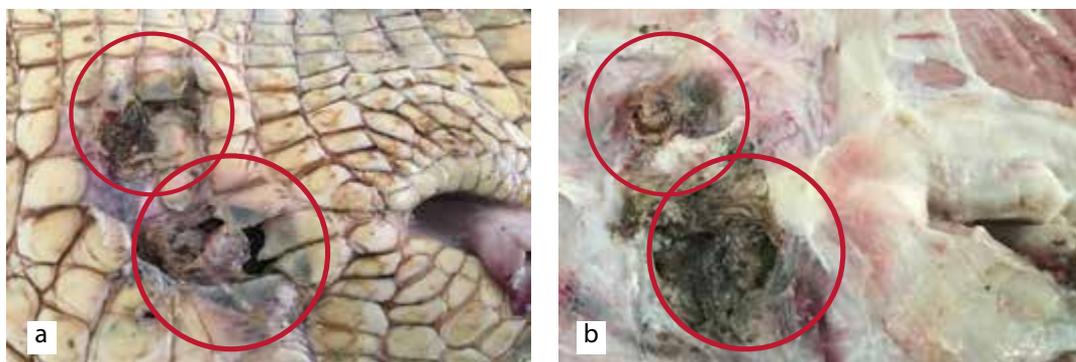
**Figure 1b.** The carcass had very bony limbs and visible spinal column.



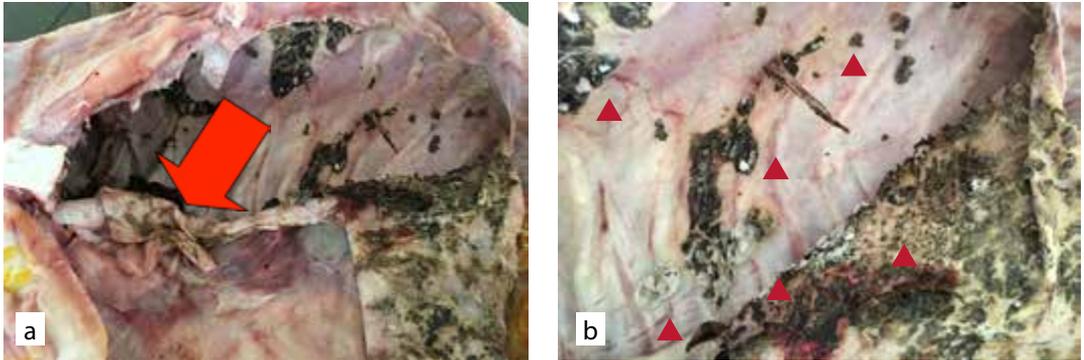
**Figure 1c.** The carcass had a very thin and left lateral kinked tail.



**Figure 2.** There was 15 cm maggot infested wound at the left quadrato/articular joint (white arrow).



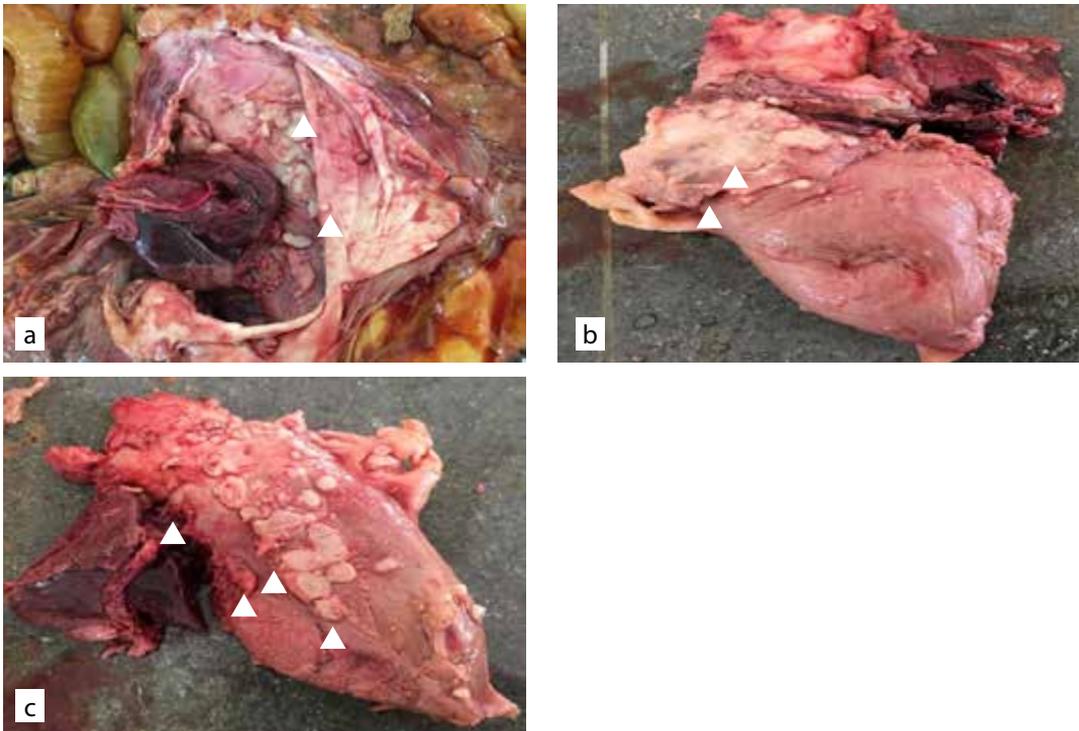
**Figure 3.** The ventral view of the pelvis (a) before deskinning, (b) after deskinning, revealed two punctured wounds located 3 cm anterior to the cloaca (circled).



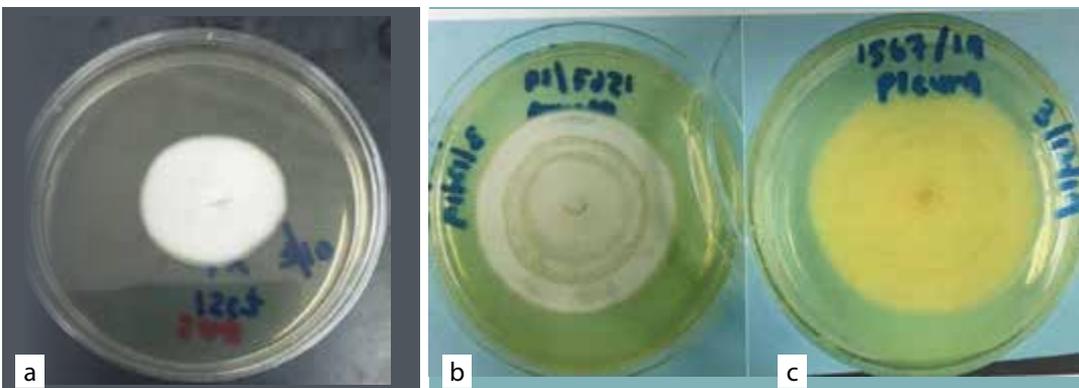
**Figure 4.** (a) The left lung had turned into brownish to blackish mushy oily substance (arrow) and (b) the left thoracic wall was filled with whitish fungal growth (arrow heads).



**Figure 5.** The right lungs had numerous whitish nodules varying in size.



**Figure 6.** (a) Severe pericarditis with presence of whitish to yellowish pus on the pericardium, (b) the base of the heart and (c) on the epicardium of the left ventricle (arrow heads).



**Figure 7.** *Geotrichum candidum* on Sabouraud Dextrose Agar (SDA), (a) fungus shown as white, fluffy colony on Day 3 post inoculation. (b) white fluffy, ringed, on Day 7 of incubation, (c) on the reverse side, Day 7 of incubation.

on Sabouraud Dextrose Agar (Figure 7a to 7c). *Klebsiella pneumoniae* and *Escherichia coli* were isolated from the thymus. The latter was also isolated from cloacal swabs.

Geotrichosis is an opportunistic mycosis that is usually succumbed by immunocompromised humans and animals (Lee *et al.*, 2011). It is common to isolate *Geotrichum* sp from lesions in reptiles (Jacobson *et al.*, 2000). However, documentation on mycotic infections in crocodiles is scarce. *Nannizziopsis* infections affecting the integumentary system (Hill *et al.*, 2019) and *Paecilomyces* infection resulting in systemic mycoses (Maslen *et al.*, 2009) have been described in captive crocodiles. Very poor body condition with noticeable thymus seen in the animal was similar to Huchzermeyer (2002) that recorded thymus is discernible in emaciated crocodiles.

In this present case, the crocodile probably endured chronic stress due to inadequate diet and overcrowding leading to immunosuppression. Later, the animal succumbed to infection.

Pal *et al.* (2013) reported two modes of geotrichosis transmission, which were inhalation and ingestion of the fungal cells. Remarkable lesions in the left lung had suggested that the animal contracted the *Geotrichum candidum* through inhalation and later contaminated the left pleural cavity, the right pleural cavity before invading the pericardial cavity. Furthermore, there is no evidence of mycosis involving the gastrointestinal tracts in this crocodile. The mixed opportunistic bacteria isolated from this animal were considered as secondary invaders because normally crocodilians have a highly effective immune system to encounter the bacterial infection (Kommanee *et al.*, 2012). To the authors' knowledge, the present case is the first to report mycosis in estuarine crocodiles caused by *G. candidum*, a ubiquitous yet potentially pathogenic, and zoonotic fungal organism.

## REFERENCES

1. Antonias N.A.B., Juffo G.D., Santos A.S., Pescador C.A., Ferreira L., and Driemeier D. (2013). *Geotrichum candidum* as a possible cause of bovine abortion, J. Vet. Diagn. Invest. 25(6):795–797.
2. Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES). Retrieved from [https://cites.org/sites/default/files/eng/app/2021/E Appendices-2021-02-14.pdf](https://cites.org/sites/default/files/eng/app/2021/E%20Appendices-2021-02-14.pdf)
3. Ellis D. (2001). *Geotrichum candidum*. Retrieved from <https://mycology.adelaide.edu.au/descriptions/hyphomycetes/geotrichum>
4. Hill A.G., Sandy J.R and Begg A. (2019). Mycotic dermatitis in juvenile freshwater crocodiles (*Crocodylus johnstoni*) caused by *Nannizziopsis crocodili*. 50(1): 225-23.
5. Huchzermeyer, F.W. (2002). Diseases of farmed crocodiles and ostriches. *Rev.Sci.tech.off.int.epiz.* 21(2):265-76.
6. Jacobson E.R., Cheatwood J.L. and Maxwell L. K. (2000). Mycotic diseases of reptiles. *Seminars in avian and exotic pet medicine.* 9(2): 94-101.
7. Lee E.J., Gabor M., Turner M., Ball M. and Gabor, L. (2011). Tonsillitis in a weaner pig associated with *Geotrichum candidum*. *J Vet Diagn Invest.* 23:175–177.
8. Lee Y., Hsu W., Lin C., Shao W. and Cheng F. (2010). Intestinal geotrichosis in a German shepherd. *Turkish J. Vet. Anim. Sci.* 34: 481-484.
9. Marcellino N., Beuvier E., Grappin R., Gueguen M. and Benson, D.R. (2001). Diversity of *Geotrichum candidum* strains isolated from traditional cheesemaking fabrications in France. *Appl Environ Microbiol.* 67(10): 4752–4759.
10. Maslen M., Whitehead J., Forsyth W.M, McCracken H., and Hocking, A.D. (2009). Systemic mycotic disease of captive crocodile hatchling (*Crocodylus porosus*) caused by *Paecilomyces lilacinus*, *Med. Mycol.* 26: 219–225.
11. Ng K.P., Soo-Hoo T. S, Koh M.T. and Kwan P.W. (1994). Disseminated *Geotrichum* infection. *Int. Med. J. Malays.* 49: 424–426.
12. Pal M., Sejra S., Sejra A. and Tesfaye S. (2013). Geotrichosis - An Opportunistic Mycosis of Humans And Animals. *Int. J. Livest. Res.* 3(2):38 Sfakianakis, A., Krasagakakis, K., Stefanidou, M., Maraki, S., Koutsopoulos, A., Kofteridis,

D., Samonis, G. and Tosca, A. 2007. Invasive cutaneous infection with *Geotrichum candidum*: sequential treatment with amphotericin B and voriconazole. *Med. Mycol.* 45: 81-84.

13. Van Dijk, C. and Thirakhupt, N. (1998). A photographic guide to snakes and other reptiles of Peninsular Malaysia, Singapore and Thailand. New Holland. Retrieved from [https://www.ecologyasia.com/verts/lizards/estuarine\\_crocodile.htm](https://www.ecologyasia.com/verts/lizards/estuarine_crocodile.htm)

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