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Pathology of proventricular tetrameriasis in a free-range chicken

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ABSTRACT: This case report aimed to investigate the aetiology of proventricular lesions and associated clinical symptoms in local free-range chickens suspected of having proventricular tetrameriasis. Proventricular tetrameriasis is a poultry disease caused by the nematode *Tetrameres* sp. In this case, 15 local free-range chickens, approximately eight months old, from Gianyar, Bali, Indonesia, exhibited symptoms of anorexia and greenish diarrhoea, resulting in the death of three chickens. One of the deceased chickens underwent thorough examinations, including anatomical pathology (PA), histopathology (HP), hemagglutinin (HA), and hemagglutinin inhibition (HI) tests for Newcastle disease virus (NDV). PA examination revealed blackish and reddish nodules on the proventriculus surface, although both HA and HI tests for NDV returned negative results. HP examination of the proventriculus tissue revealed cross-sections of *Tetrameres* sp. nematodes with pseudocoeloms filled with bright eosinophilic fluids. Additionally, ectasia of the proventricular glands, with compression atrophy and mild inflammation, was observed.

Keywords:
chicken, gross pathology, histopathology, proventriculitis, *Tetrameres sp.*

■ INTRODUCTION

Tetrameres sp. is a nematode belonging to the suborder Spirurida, primarily targeting poultry, with a predilection for the proventriculus. Infection with *Tetrameres* sp. can result in severe proventriculitis and a reduction in digestive secretions, leading to clinical symptoms such as weakness, emaciation, and anaemia (Gao *et al.* 2022). Documented cases of *Tetrameres* sp. infections have been reported in chickens and turkeys in Nigeria (Kamani *et al*. 2010) and in chickens in India (Govindan & Annamalai 2019). In Bali, *Tetrameres* sp. infection has been reported in ducks (12%, n=50) and free-range chickens (33.6%, n=110) (Yulianda *et al*. 2023; Chandra *et al*. 2017). Although the histopathological features of proventricular tetrameriasis have been described in ducks in Bali (Yulianda *et al*. 2023), similar reports in free-range chickens remain scarce (Chandra *et al*. 2017). This case report aims to observe the clinical presentation, anatomical pathology, and histopathology of *Tetrameres* sp. infection in free-range chickens.

■ CASES

Signalment and Case History: Fifteen free-range chickens, approximately eight weeks old, from Gianyar, Bali, Indonesia, presented with clinical signs of anorexia and greenish diarrhoea. Despite receiving vitamin supplementation, three affected chickens eventually dead from the illness. To ascertain the precise cause of death, a comprehensive necropsy was meticulously conducted on one of the deceased chickens at the Veterinary Pathology Laboratory, Faculty of Veterinary Medicine, Udayana University. **Gross Pathology Findings**: Necropsy revealed blackish nodules on the surface of the proventriculus, suggestive of a spirurid nematode infection (Figure 1). The proventriculus appeared thickened, and in addition to the blackish nodules, reddish spots resembling petechial haemorrhages were observed on the surface, initially suggesting a presumptive diagnosis of Newcastle Disease. **Ancillary Tests**: Tissue homogenates from the deceased chicken were rigorously tested for Newcastle disease virus using hemagglutination (HA) and hemagglutination inhibition (HI) assays, all of which returned negative results, ruling out the initial presumptive diagnosis.

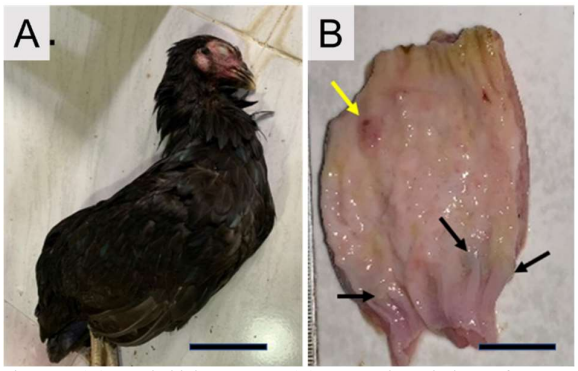

Figure 1. (A) Dead chicken carcass (B) Anatomic pathology of proventricular tetrameriasis. The proventriculus showed black-coloured nodules (black arrow) and a reddish spot (yellow arrow). Bar 1 cm.

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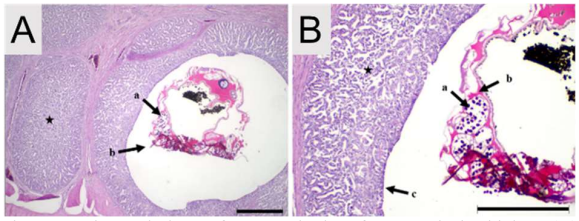

Figure 2. Histopathology of proventricular of tetrameriasis chicken. (A) Cross-section of *Tetrameres sp.* (a). Submucosal glands are markedly dilated (b). The proventricular glands inflamed (star). Bar 100 µm. (B) These parasites (a) can be identified by the eosinophilic pseudocoelomic fluid (b). Desquamation of glandular epithelium (c) with atrophied glandular acini (star). Bar 50 µm. Hematoxylin-Eosin (HE).

Histopathological Findings: Histopathological examination of the proventriculus revealed cross-sections of helminths within the glands. The parasites were identified as *Tetrameres* sp., characterized by their spherical shape and pseudocoelom containing abundant homogeneous eosinophilic fluid (Figure 2). The proventriculus also showed lymphocytic infiltration, with rarely heterophils present. **Morphologic Diagnosis**: Proventricular gland ectasia with compression atrophy, intraglandular spirurid nematodes, and mild necrotizing proventriculitis. **Etiologic Diagnosis**: Proventricular tetrameriasis.

■ RESULT AND DISCUSSION

Tetrameres sp. is a parasitic nematode infecting various bird species, including chickens, ducks, grouses, pigeons, turkeys, and quails, regardless of age. Transmission occurs mainly through intermediate hosts like grasshoppers, amphipods, and cockroaches (Taylor *et al*. 2016). Chandra *et al*. (2017) noted that free-range chickens infected with *Tetrameres* sp. are typically three to five months old. Additionally, Fink *et al*. (2005) observed that *Tetrameres americana* prevalence is higher in younger chickens (less than two months old) compared to growers (2-8 months) and adults (over eight months). Younger chickens face greater infection risks and often experience more severe outcomes than adults (Taylor *et al*. 2016).

Anatomical pathology examination of the chicken cadaver revealed black and red nodules in the proventriculus, with reddish spots indicative of gravid *Tetrameres* sp. females on the serosal surface. These findings align with those of Kamani *et al*. (2010), Govindan & Annamalai (2019), and Chandra *et al*. (2017). Histopathological examination showed worm cross-sections in the proventricular gland, similar to *T. fissipara* infections in ducks (Kamil *et al.* 2011). Unlike Megha *et al*. (2022), who reported numerous embryonated eggs with acidophilic shells and basophilic larvae, our case exhibited desquamation of glandular epithelium and inflammation with atrophy of glandular acini due to worms in the proventricular glands. These pathological changes are consistent with findings in ducks by Megha *et al.* (2022), Kamil *et al*. (2011), and Yulianda *et al.* (2023).

Tetrameres sp. is easily recognizable due to extreme sexual dimorphism, with male and female forms exhibiting distinct red coloration. Female worms are round (globular), while males are elongated, as observed in most nematodes (Taylor *et al.* 2016). *Tetrameres sp.* can infect poultry through the ingestion of intermediate hosts. Adult worms develop in the proventriculus by feeding on blood and becoming engorged and gravid. The primary cause of death is often attributed to worms embedded in the proventricular serosa (Kamani *et al.* 2010). Apart from blood feeding, female *Tetrameres sp.* worms can locally erode the glands in the proventriculus (Taylor *et al.* 2016). The specific species of *Tetrameres* infecting chickens in this case report remains unidentified, warranting polymerase chain reaction and sequencing tests for identification. Known species of *Tetrameres* infecting chickens include *T. americana*, *T. fissispina*, *T. confuse*, and *T. mohtedai* (Taylor *et al.* 2016). However, the species of *Tetrameres* infecting poultry in Indonesia remains unknown.

■ CONCLUSION

Based on signalment, history, as well as the findings from PA, HP, HA, and HI examination, it was concluded that the chicken in the case was afflicted with proventricular tetrameriasis.

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