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Short Communication

Pathological Findings of *Eimeria tenella* in Desi Chickens with Morphometric Analysis of Oocysts

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Abstract

Five numbers of three weeks old dead desi chickens were brought to the Department of Veterinary Pathology, Veterinary College and Research Institute, Tirunelveli for postmortem examination. External examination revealed emaciation with ruffled feathers and pale mucous membrane. On internal examination, liver showed multifocal pale areas, lung was congested and caecum lumen contained clotted blood with multifocal ecchymotic hemorrhages. Cytological examination of intestinal scrapings revealed the presence of numerous unsporulated oocysts appeared as oval to round shaped with double layered wall. Histopathological examination of internal organs showed, mild degeneration of hepatocytes and diphtheritic enteritis along with presence of numerous Eimeria tenella coccidial oocysts studded within the mucous membrane. The oocysts morphometric analysis was carried out in 40x magnification of light microscope. The values of Eimeria tenella oocysts like breadth, length and shape index were: 24.48µm, 30.84µm and 1.25µm, respectively. Based on cytological, histopathological and morphometry analysis of oocysts, the case was diagnosed as caecal coccidiosis caused by Eimeria tenella. The flock was treated with Amprolium hydrochloride 20% in drinking water for a period of 4 days and was advised to change the bedding material.

Keywords: Coccidiosis, *Eimeria tenella*, Oocysts, Histopathology.

Introduction

Coccidiosis in poultry is a most recognized parasitic disease caused by apicomplexan parasites of the genus *Eimeria* (Mares *et al.,* 2023). Chickens can be affected with these species which includes *Eimeria acervulina, Eimeria brunetti, Eimeria praecox, Eimeria maxima, Eimeria mitis, Eimeria necatrix, and Eimeria tenella* with different clinical form of pathological effects (Williams *et al.,* 2005). Among these coccidian parasites *E. tenella* and *E. necatrix* are regarded as the most pathogenic to young chickens (Vijayalingam *et al.,* 2019: Aiello and Moses, 2016). *Eimeria* species are multiply in the epithelial cells of intestinal villi and crypts, which are host and sites specific (Varghese, 2004). Sexual and asexual reproduction of each species grows within the intestinal epithelium of its definitive host and producing oocysts and that are expelled in the feces (Kaufman, 1999). The clinical symptoms include dullness, enteritis, bloody droppings (Samal *et al.,* 2017) and loss of body weight. The mode of infection may be due to contact with the droppings of the bird or contaminated food and water. The majority of *Eimeria* species have coccidia oocysts with a variable morphology. The majorities of oocysts from *Eimeria* are ovoid in form and have a rigid cell wall. In comparison to other *Eimeria* species, *E. maxima* (30.5 x 20.7 μm) is the largest, while *E. mivati* (15.6 x 13.4 μm) and *E. mitis* (15.6 x 14.2 μm) are the smallest. While *E. necatrix* is oblong, *E. tennela, E. maxima, E. acervulina, E. hagani, and E. brunetti* are ovoid (Clark and Blake, 2012). Unsporulated oocysts are not infectious but sporulated oocysts are infective stages and spread the infection. Sporulated oocysts are able to survive in environment for 602 days (Lal *et al.,* 2009). The present study aimed to combine cytological, histopathological and oocysts morphometric methods so as to identify *Eimeria* species in desi chickens.

Materials and Methods

Five numbers of three weeks old desi chickens were brought to the Department of Veterinary Pathology, Veterinary College and Research Institute, Tirunelveli for postmortem examination. The history revealed that, the farm was holding around 2000 birds maintained in deep litter system and mortality pattern of five to six birds daily over a period of one week. The owner reported that the dead birds exhibited the clinical signs like ruffled feather, anorexia and occasionally blood droppings. Complete postmortem examination was conducted as per standard procedure. The impression smears were taken from liver and lungs and stained with Leishman's stain intended for cytological examination. Intestinal scrapings were taken from different sites of intestinal epithelium used for wet mount examination. The morphometry was carried out in 40x magnification of light microscope by taking the mean values of measurements of at least 100 oocysts of each sample from different birds. (Venkateswara *et al.,* 2012). Internal organs were collected and preserved in 10% formalin for histopathological examination. The Paraffin embedded tissue section of 3μm thickness were cut and stained with Hematoxylin and Eosin as per the standard procedures.

Results and Discussion

Poultry coccidiosis is economically significant problem in commercial poultry production and to a great extent affected due to mortality and worldwide losses it can cause more than 3$ billion per year (Dolloul *et al.*, 2006). On post mortem examination, the carcasses were emaciated with ruffled feather, pale mucous membrane with clinical history of bloody drooping. Similar findings were also described by previous report by (Olabode *et al.*, 2020). In our report macroscopically, liver showed multifocal pale coloured areas and Lung was congested. Small intestinal wall thickened, few patchy of congestion and yellow coloured mucoid content. Caecum lumen contained clotted blood with multifocal ecchymotic haemorrhages and multifocal necroses (Fig. 1) were noticed on the mucous membrane. Caecal tonsils showed mild enlargement and petechial haemorrhages. Similar types of lesions were recorded in many reports like thickened intestinal wall, blood-tinged exudates, petechial hemorrhages and necrosis in intestine and mucous profuse bleeding in the caecum (McDougald and Hu, 2001). Cytological examination of intestinal scrapings revealed the presence of numerous unsporulated coccidial oocysts (Fig. 2) appeared as oval shape with double layered wall. This is in agreement with previous report of (Samal *et al.,* 2017). Histopathological examination of intestine especially caecal mucous membrane revealed necrosis (Fig. 3), infiltration of inflammatory cells within the sub mucosa along with the presence of numerous *Eimeria tenella* oocysts which were studded within the mucous membranes (Fig. 4). The death of the chickens might be due to heavy blood loss and necrosis of intestinal epithelium which results poor absorption of nutrients. These findings were in accordance with the earlier work (Olabode *et al.,* 2020). Morphometric measurement of oocysts performed in unsporulated oocysts, were as other earlier studies (Mares *et al.,* 2023; Samal *et al.,* 2012) used sporulated oocysts. In our study oocysts were appeared as broad and ovoid with double-layered walls. The dimensions of the unsporulated oocysts are 22 (20–24) μm in length and 15 (17–20) μm in width and 1.25 μm shape index. The morphometric analysis of oocysts detected in our

study was slightly different, with those described in other studies used sporulated oocysts. We also analyzed the morphometry of oocysts in haematoxylin and eosin sections (Fig, 5 & 6), the values were (20–24) µm in length and 15 (17–20) µm in width and 1.25 µm shape index. Morphometry of oocysts aids for specific identification of *Eimeria* species. The slight differences in oocyst measurement leads to inaccurate conclusions about species, which could be explained by variations in the parasites or bird's metabolisms or even overlaps in the form of morphometric indices (Haug *et al.*, 2008). Thus, further molecular study is important role to verifying species differentiation among *Eimeria sp*.

Fig. 1. Desi chickens - Presences of necrosis and haemorrhages in caceum

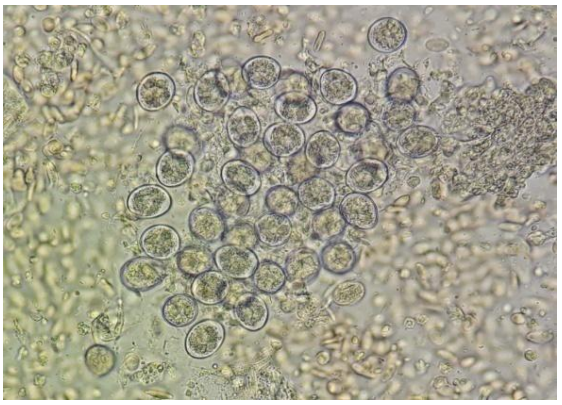

Fig. 2. Ovoid to round shaped *Eimeria tenella* oocysts – Intestinal scraping – wet mount exam

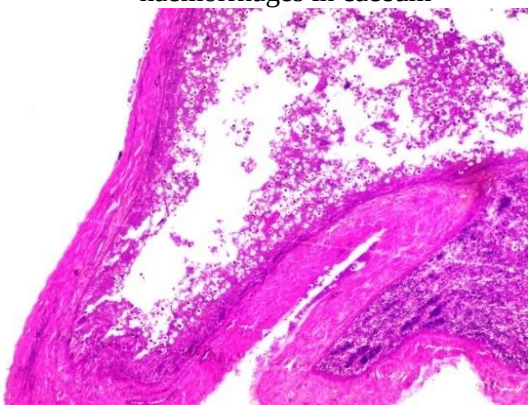

Fig. 3. Necrosis of caecal mucous membrane – Numerous oocysts - H&E x40

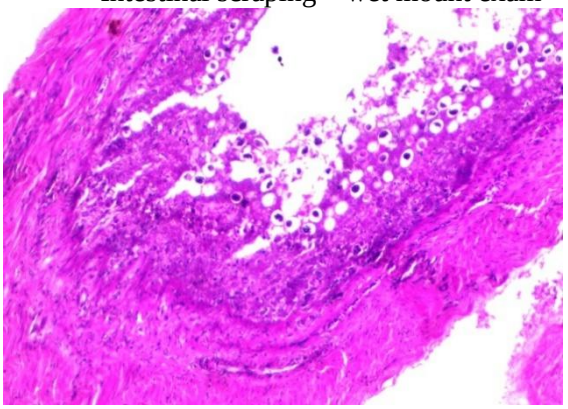

Fig. 4. Presences of *Eimeria tenella* oocysts within the sub mucosa - H&E x100

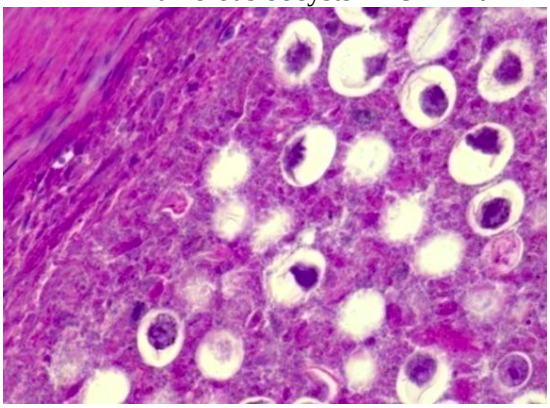

Fig. 5. *Eimeria tenella* oocysts – Caecum - H&E x400

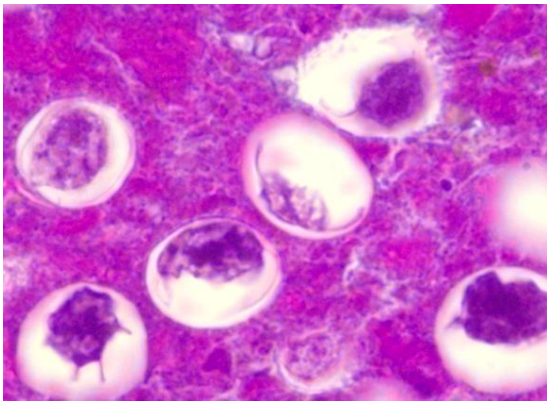

Fig. 6. Round to oval shaped *Eimeria tenella* oocysts – Caecum - H&E x1000

Conclusion

Based on cytological, histopathological and morphometry analysis of oocysts, the case was diagnosed as caecal coccidiosis caused by *Eimeria tenella*. The flock was treated with amprolium hydrochloride 20% in drinking water for a period of 4 days and also advised to maintain the proper biosecurity measures and hygiene.

Contribution by Authors

All the authors contributed equally to writing the manuscript. The final manuscript was read by all authors and consented to publication.

Conflict of Interests

There is no conflict of interest.

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