

***Hexamita* infection associated with diarrhoea and stunting in native turkey pouls**

Dezfoulian, O.^{1*}, Gharagozlu, M.J.² and Rahbari, S.²

¹Section of Pathology, School of Veterinary Medicine, University of Lorestan, Khorramabad-Iran

²Department of Pathobiology, Faculty of Veterinary Medicine, University of Tehran, Tehran-Iran

²Department of Pathobiology, Section of Parasitology, Faculty of Veterinary Medicine, University of Tehran, Tehran-Iran

*Corresponding author email: omidvete@yahoo.com

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Abstract. *Hexamita* sp. was identified in 8 cases of sixty (13.3%) stunted diarrhoeic 1-12-week old native turkey pouls. Large numbers of the flagellated parasites were found within the dilated crypts of Lieberkühn of ileum and jejunum. In TEM studies on the parasagittal section of the parasite, elongated flagellated binuclear parasite was found. The morphological characteristics of the seen protozoan are similar with those described for *Hexamita meleagridis*. The parasite could be considered as an intestinal pathogenic protozoan parasite, causing stunting and diarrhoea in turkeys in Iran.

INTRODUCTION

Hexamita parasite is a pathogenic flagellated protozoan which infects various species of birds including turkey (Du-Whan, 1975; Stepkowski, 1979; Stoikov, 1985), pheasant (Swarbrick, 1985; Swarbrick, 1990; Milne et al. 1997), chukar partridge (Cooper et al., 2004), Crane (Ippen et al., 1980), pigeon (Stepkowski 1979; Reece et al., 1992), quail (Mc Dougald, 2008) and peafowl (Mc Dougald, 2008). Several reports from natural outbreaks of hexamitiasis indicate that the protozoan is a potential pathogen and the presence of the organisms in the gut could be associated with clinical manifestation (Ippen et al., 1980; Swarbrick, 1990; Reece et al., 1992; Cooper et al., 2004; Mc Dougald, 2008).

Two species of *Hexamita* identified in birds include *Hexamita meleagridis* which was reported in turkeys, pheasant and chukar partridge (Du-Whan, 1975; Stoikov, 1985; Cooper et al., 2004; Mc Dougald, 2008), and *Hexamita columbae* which is

infective to pigeon (Reece et al., 1992; Mc Dougald, 2008). Hexamitiasis has been reported from Netherland (Reece et al., 1992), Germany (Ippen et al., 1980; Mc Dougald, 2008), Scotland (Mc Dougald, 2008), United Kingdom (Swarbrick, 1985; Swarbrick, 1990), Australia (Zwart & Hooimeijer, 1985), Bulgaria (Stoikov, 1985), Korea (Du-Whan, 1975), Canada (Mc Dougald, 2008), and United States (Cooper et al., 2004; Mc Dougald, 2008). In the present study, *Hexamita* infection is reported in diseased turkey pouls. By the best knowledge of the authors, this is the first report of *Hexamita* infection in turkeys from Iran.

MATERIALS AND METHODS

Sixty 1-12 week-old native turkey pouls (*Meleagris gallopavo*) showing clinical signs of unthriftiness and chronic diarrhoea obtained from rural area of Mazandaran (north province of Iran) and Lorestan

(south-west province of Iran) were examined for the causative agent of the disease. The native turkeys were kept as small flocks generally consisting of chickens, turkeys and waterfowls and they were kept under free range condition. The clinical manifestations of the affected birds were recorded and they were sacrificed. The tissue samples taken from duodenum, jejunum, ileum, caecum, colon, cloacae and bursa of Fabricius were fixed in 10% buffered formalin (Ph = 7.2). The formalin-fixed tissues were processed in a tissue processor, paraffin blocks were made, 5-6 µm thick tissue sections were stained with Harris hematoxylin and eosin (H&E) method. Selected parts of the formalin fixed segments were post-fixed in 2.5% glutaraldehyde and processed through standard method for transmission electron microscopy (TEM). The ultra - thin sections stained with uranyl-acetate and lead citrate were examined under a Philips 208 S transmission electron microscopy.

RESULTS

Clinical findings: Watery diarrhoea, emaciation, growth retardation were observed in the affected birds.

Pathology: The carcasses of the poult were poor and thin, the musculature of the pectoral muscle was dehydrated, small intestine was distended and frothy yellow watery fluid were noticed within the affected areas of intestine. Histological examination of the sections stained with hematoxylin and eosin as well as toluidine blue, showed large numbers of *Hexamita* protozoan parasites appearing as elongated conical shape within the dilated crypts of Lieberkühn of the jejunum and ileum (Figs. 1 & 2). The binucleated bodies were prominent only in toluidine blue staining method. Hyperplasia of crypt epithelium and increased in lamina propria inflammatory cells were seen in the affected tissues. *Hexamita* organisms were found in 8 cases of the clinically affected birds 60 (13.3%). In TEM studies, the

elongated, conical, binuclear flagellated *Hexamita* with a prominent recurrent flagellum and a large cytostome was identified (Fig. 3).

DISCUSSION

The present study indicates that *Hexamita* infection is present in native turkeys in Iran, and it is associated with clinical signs of diarrhoea, emaciation and growth retardation. The clinical manifestations described in this study are compatible to those that have been reported by other workers (Du-Whan, 1975; Stepkowski, 1979; Stoikov, 1985; Swarbrick, 1985; Swarbrick, 1990; Reece *et al.*, 1992; Cooper *et al.*, 2004; Mc Dougald, 2008). The clinical signs and pathological changes in the intestine associated with hexamitiasis in turkeys (Du-Whan, 1975; Stepkowski, 1979; Stoikov, 1985; Mc Dougald, 2008), pheasants (Swarbrick, 1985; Swarbrick, 1990; Milne *et al.*, 1997; Mc Dougald, 2008), pigeons (Stepkowski, 1979; Reece *et al.*, 1992) cranes (Ippen *et al.*, 1980) and Chukar partridges (Cooper *et al.*, 2004) indicate that the protozoan is a potential pathogen in birds. The parasite causes detrimental effects on intestinal mucosa (Ippen *et al.*, 1980; Swarbrick, 1985; Swarbrick, 1990; Reece *et al.*, 1992; Cooper *et al.*, 2004; Mc Dougald, 2008). The mucosal damage caused by the protozoan parasite may have allowed bacterial entry resulting in bacteraemia and organ colonization (Mc Dougald, 2008). The macroscopic and microscopic findings including poor carcasses, yellowish watery contents and distention in the small intestine, catarrhal enteritis with dilatation and hyperplasia of affected crypts of Lieberkühn and presence of binucleated conical *Hexamita* protozoan within the dilated crypts are similar to the findings reported by other workers (Du-Whan, 1975; Stepkowski, 1979; Stoikov, 1985; Swarbrick, 1985; Mc Dougald, 2008; Zwart Hooimeijer, 1985). Two species of *Hexamita* have been reported to be causative agents of

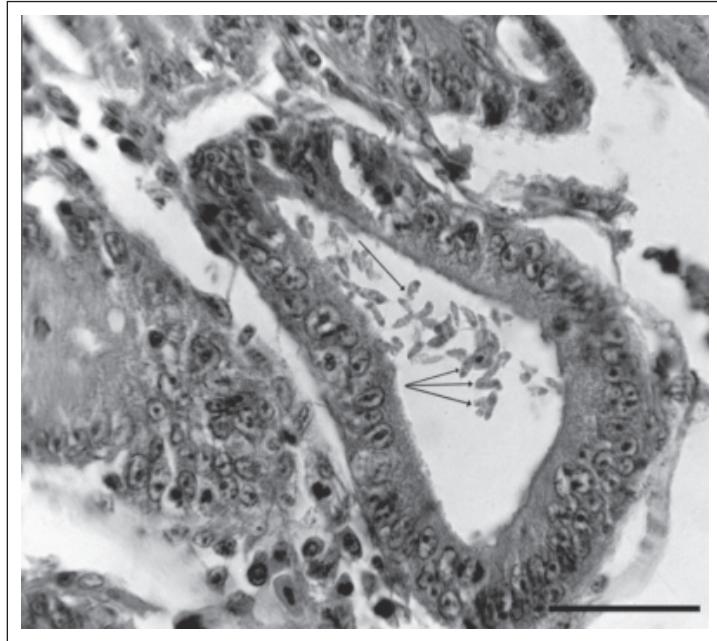


Figure 1. Large numbers of elongated *Hexamita* organisms within the crypt of liberkuhn (Arrows). HE. Bar = 40 μ m

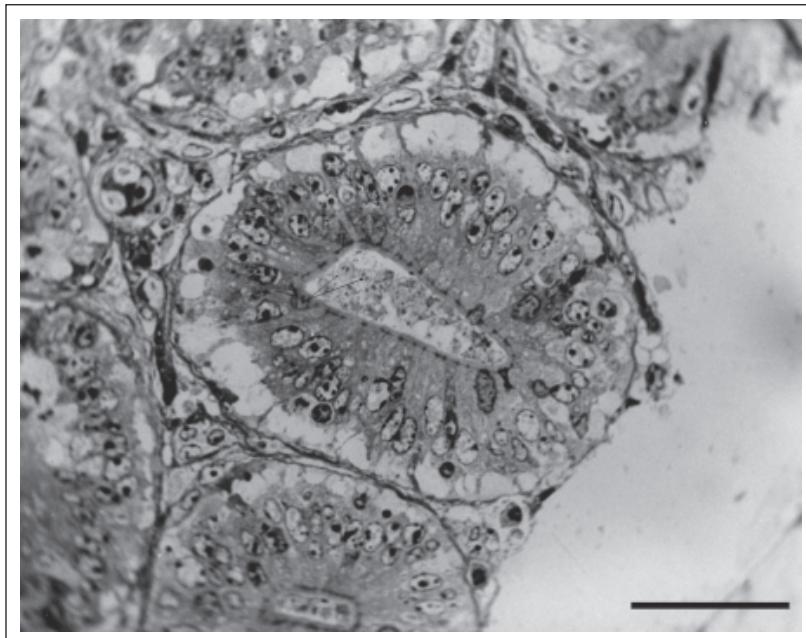


Figure 2. A plastic section from the small intestine of the infected turkey poult. Elongated, conical shaped, binucleated *Hexamita* parasite within the crypt of liberkuhn (Arrows). TB. Bar = 40 μ m

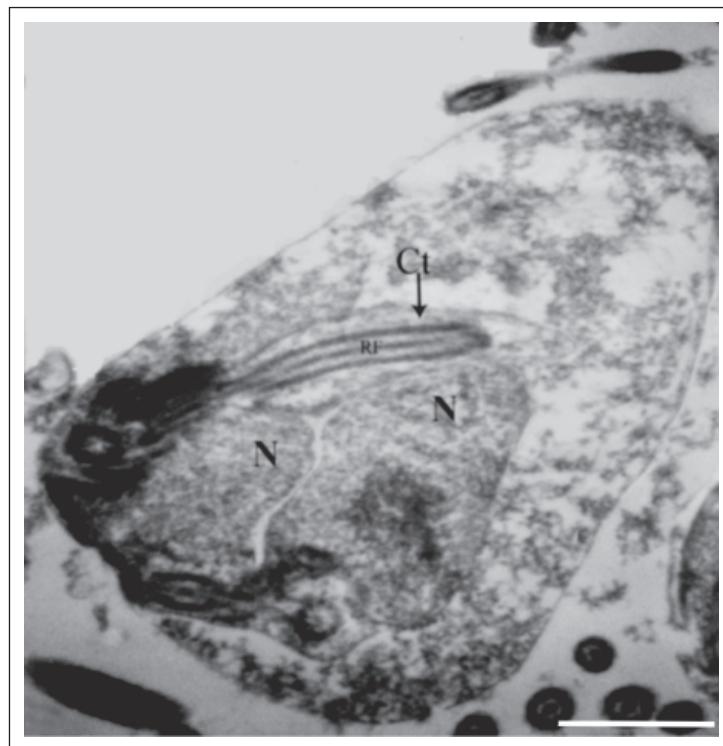


Figure 3. Transmission electron micrograph of *Hexamita meleagridis* trophozoite Within the intestinal crypt; N (nucleus), Ct (cytostome), R Fl (recurrent flagellum) Fl (flagella). Bar=1.5 μ m

hexamitiasis in different species of birds, namely *H. meleagridis* in turkeys and *H. columbae* in pigeons. Based on species of birds infected and morphological characteristics of the parasite, it is most likely that the species of the *Hexamita* reported here is *H. meleagridis* which has been described previously by others (Swarbrick, 1985; 1990; Cooper *et al.*, 2004; Mc Dougald, 2008). Slender elongated binuclear trophozoite, multiple flagella including a prominent recurrent flagellum and a large cytostome were morphological findings in the present study which are similar with *H. meleagridis*. Regarding the present and other reports of the natural infection, young birds are the most susceptible to the infection and it is almost always accompanied by clinical manifestations of poor growth, emaciation, diarrhoea and mortality among the

affected birds (Stepkowski, 1979; Ippen *et al.*, 1985; Stoikov, 1985; Swarbrick, 1985; 1990; Reece *et al.*, 1992; Milne *et al.*, 1997; Cooper *et al.*, 2004; Mc Dougald, 2008). The young birds that survived infection grow to become asymptomatic reservoir adult birds (Swarbrick, 1985; 1990; Cooper *et al.*, 2004; Mc Dougald, 2008). Nevertheless, the infected native turkey pouls and other asymptomatic birds harboring *Hexamita* parasite could be potential sources of the disease agent not only for turkey industries but also for susceptible free living birds and birds kept under zoo condition. There is no effective treatment for the infection (Mc Dougald, 2008), although Butymorate, Chlortetracycline, combination of Furaltadone and Tetracycline, Furazolidone and Ranidazole could be prescribed (Swarbrick, 1990; Cooper *et al.*, 2004; Reece *et al.*, 1992; Mc Dougald, 2008). To

control hexamitiasis in birds, hygienic measures including control of house flies and other arthropods and inclusion of Dimetridazole at 125 ppm to 150 ppm are recommended (Swarbrick, 1990; Cooper *et al.*, 2004; Mc Dougald, 2008).

It is concluded that *Hexamita* is one of the intestinal protozoan infection in native turkeys of Iran and the infected turkeys may have an important role in the epidemiology of the disease.

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