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INTESTINAL PROTOZOA OF TURKEYS IN SHARKIA GOVERNORATE

N.M . EL-SAYED* and AZZA, M . RAEF **

Dept. of Parasitology , Fac. Vet. Med., Zagazig University*
Animal health research institute, Agricultral Research center, Dokki, Giza , Egypt**

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SUMMARY

Fifty intestinal tract of domesticated turkeys were collected from private poultry markets in Zagazig city,Sharkia province for detection of protozoan parasites . Eleven out of fifty (22 %) intestinal tract were infected with protozoan parasites. Seven of the examined samples (14%) were infected with three species of *Eimeria* ; *E. adenoeides* 7/50 (14 %); *E.meleagrimitis* 5/50 (10%) and *E.innocua* 4/50 (8%) . Three of the examined samples (6%) were infected with *Cryptosporidium meleagridis* oocysts. Mixed infection with Trichomonads (*Tetratrichomonas gallinarum* and *Tritrichomonas eberthi*) were detected in one of the examined samples (2%). Experimental infection of white albino rats with 10^5 of *C. meleagridis* oocysts revealed the presence of oocysts in faeces of infected rats 4 days post-infection. Histopathological examination of different organs of infected rats at 4 days post-infection showed endogenous developmental stages (trophozoites and schizonts) of *C. meleagridis* at brush border of tracheal epithelium. The characteristics morphological features for the detected species were illustrated with photographs .

INTRODUCTION

Poultry have been on the earth for over 150 million years, dating back to the original wild jungle fowl . Now turkeys ,ducks, geese, pheasants, pigeons, peafowl and guinea fowl chickens are included in the list of species under the general term poultry. Poultry provide humans with companionship, food and fiber in the form of eggs, meat and feathers. Turkeys are intensively reared for their meat. The later is low in fat and high in protein.

Protozoa of two major groups are responsible for economic damage to poultry. These are coccidia

and the flagellates including *Histomanas* and *Trichomnas* (McDougald, 1997) . Coccidiosis is a disease of universal importance in poultry production . It is common in turkey , but is often unrecognized because the lesion are less spectacular than those in chicken. It has long been consider a disease of young turkey poult , however older turkey that had never been exposed to coccidia have also shown to be susceptible to infection (Morehouse,1949 and Edgar,1986). Several species of *Eimeria ;E. adenoeides, E. gallopavonis E. meleagridis, E.meleagrimitis, E. dispersa, E . innocua* and *E.subrotunda*; are responsible for coccidiosis in turkey (Moore,1954; Pellardy,1974 and Mcdougald,1997). Significant reduction in weight gain , watery mucoid diarrhea, ruffled feather and mortality are common in *E.adennoides , E. meleagrimitis, E. gallopavonis* , but other species cause reduced weight gain ,dehydration , malabsorption. of specific nutrients,secondary infection and may potentiate other diseases (McDougald, 1997).

Cryptosporidiosis have been reported in several species of birds including turkey , chicken, duck, geese , quail, pheasant and wide varieties of wild and captive birds .Two species of Cryptosporidium which infect birds are accepted; *C.baileyi* in chicken and *C.meleagridis* in turkey (Fayer et al.,1997) . It is usually manifested as a respiratory or enteric diseases (Goodwin, 1989 and Lindsay & Blagburn, 1990). Intestinal cryptosp- -oridiosis due to *C.meleagridis* was associated with diarrhea ,unthriftiness and moderate morbidity, also *C.meleagridis* has been incriminated as the causative agent in turkey with respiratory diseases and sinusitis(Darabus,1997) .On the other hand, *C.meleagridis* has been linked with several investigators to intestinal and respiratory cryptosporodiosis in human (Morgan et al ,1999; Tizipori &Widmer 2000; Carreno et al.,2001; Tizipori &Ward,2002 and Akiyoshi et al., 2003).

Flagellates are common in the intestinal tract of birds , but few are responsible for serious diseases and economic loss. There are several species of trichomonads infect commercial and noncommercial birds . Most infections are symptomless although some highly virulent strains are known. These include *Trichomonas gallinae, Tetrtatrichomonas gallinarum, Chilomastix gallinarum, Cochlosoma anatis and Hexamita meleagridis.* Little is known about these parasites as there has been no research on theses organism for many years (McDougald,1997). *Trichomonas gallinae, Tritri chomonas eberthi , Tetratrichomonas gallinarum* are the causative agents of Tichomoniasis in turkey BonDurant and Honigberg (1994).

Many investigators at different parts of the worlds work on protozoan parasites of turkey (Moore, 1954 ; Bond,1967 ; Edger,1986 ; Ozer et al. 1990; Morgan et al ,1999 ;Carreno et al., 2001 ; Tizipori&Ward, 2002 and Akiyoshi et al.,2003).while, little is known about that parasites in Egypt (Khalifa,1978 and Bassioni et al., 1979). So, this study

aimed to investigate the intestinal protozoa of turkey in Sharkia province; dealing with their prevalence, morphological identification of their different stages and the possibility of infection of mammals with turkey parasites

MATERIALS AND METHODS

Fifty intestinal tract of domesticated turkeys were collected from private poultry markets in Zagazig city, Sharkia province . Each part of gastrointestinal tract was dissected separately in petri dishes . The content of each part was examined by direct smear or floatation concentration technique for detection of oocysts. Also, the contents and mucosal scrapings of caeca were examined directly or stained with Giemsa stain for detection of Trichomonads (Levine, 1985).

The coccidial oocysts in the positive cases were washed several times in physiological saline then sporulated in 2.5 % potassium dichromate solution at room temperature (Williams, 1969) for identification of oocysts. Samples from infected intestine were taken for detection of the developmental stages of coccidia in histopathological section . *Cryptosporidium* oocysts from scraping of small intestine were concentrated and counted by using haemocytometer slide (Kuczynka&Daniel, 1999). Group of three albino rats of two weeks old was infected orally each with 10^5 Cryptosporidium oocysts while the other group of rats was kept as non infected control . Faecal samples from both groups were examined daily for detection of oocysts . Samples from different organs (intestine, liver , trachea and lung) of freshly killed rats of both groups were taken at 4 days post-infection for histopathological examination (Carleton et al., 1967).

RESULTS

Eleven out of fifty (22 %) intestinal tracts of domesticated turkeys were infected with protozoan parasites. Seven of the examined samples (14 %) were infected with three species of *Eimeria ; E. adenoeides* 7/50(14%); *E.meleagrimitis* 5/50 (10%) *E.innocua* 4/50 (8%). Mixed infection with *E. adenoeides ; E.meleagrimitis ; E.innocua* was detected in four samples while double infection with *E. adenoeides* and *E.meleagrimitis* was found in two samples .Three of the examined samples (6%) were infected with *Cryptosporidium meleagridis* oocysts .Mixed infection with Trichomonads (*Tetratrichomonas gallinarum and Tritrichomonas eberthi*) were detected in one of the examined samples (2%) . The detected protozoan parasites were morphologically identified as following :

1- *Eimeria adenoeides* (Moore and Brown , 1951). Fig. 1-3

The oocyst is ellipsoidal or elliptical in shape with smooth wall and prominent micropyle. It measures 19-30 x 12.7-17.5 (25.8 x15.7)um . One or two polar granules appear as refractile bodies inside the sporulated oocyst while the oocyst residual body is absent. The sporocyst is elongated with stieda and residual bodies. The sporulation time was 24 hrs at room temperature. Spherical shaped trophozoite with eccentric nucleus and schizont were observed in section of caecum (Fig. 8-9).

2- *Eimeria meleagrimitis* (Tyzzer, 1929). Fig. 4-5

The oocyst is ovoid with double contoured wall and measures 17-22 x 22.5 -19.3x16.5 um. The micropyle , oocyst residuum are absent while the polar granule is present in the sporulated oocyst . The sporocyst is oval with stieda body at pointed end and contain residual body .The sporozoite contain colourless globule at the broad end .The sporulation time was 24-72 hrs at room temperature.

3- *Eimeria innocua* (Moore and Brown, 1952). Fig. 6-7

The oocyst is round to sub-spherical with double contoured wall measuring 22.2x21 um. The micropyle, oocyst residuum and polar granule are absent . The sporocyst is ovoid with stieda and contain residual body .The sporozoite contains colourless globule at the broad end .The sporulation time was 24 hrs at room temperature

4- *Cryptosporidium meleagridis* (Slavin , 1955). Fig.10

The oocysts is ovoid or spherical with thin wall and measures 4-5.5 (4.7) um . It has a cytoplasm of a spumy structure and contains four sporozoites

Experimental infection of albino rats with 10^5 *C.meleagridis* oocysts revealed the presence of oocysts in faeces of infected rats 4 days post-infection Histopathological examination of different organs of infected rats killed after detection of oocysts in faeces (4 days post-infection) showed developmental stages (trophozoites and schizonts) of *C.meleagridis* at brush border of tracheal epithelium (Fig.11). Neither oocysts nor developmental stages were detected in the faeces or examined tissues of control group of rats respectively.

5- *Tetratrichomonas gallinarum* (Martin and Robertson, 1911) Fig. 12-13

It was detected in mucosal srcap of caeca . It is ellipsoidal or oval in shape and measures 7-15x 4-9 (12 x 5.6) um. The four anterior flagella have tendency to be arranged in two groups.They are sub-

equal within each group. The undulating membrane is well developed and about as long as the body which continue as free flagellum beyond the posterior end of the organism. The costa as long as the undulating membrane is somewhat thickened and usually accompanied by row of costal granules. The Axostyle varies from being slender to rather stout with its terminal part projecting for some distance beyond the posterior body surface . The nucleus is ellipsoidal , ovoid or spheroid while the para-basal body is discoid.

6-*Tritrichomonas eberthi* (Martin and Robertson, 1911) Fig. 14- 15

It was recovered from caecal scrap . It varies in shape but typical ones tend to be ovoid or slightly elongated and measures 8.6-14 x4-9.8 (10.4x 6.5) um . There are typically three equal or subequal anterior flagella . The undulating membrane is well developed and supported by a relatively heavy costa that tapers gradually toward both ends . The recurrent flagellum extends beyond the posterior end for a distance equaling one half of the length of the organism . The elongate or ovoid nucleus is located a short distance posterior to the anterior surface of the body . The projecting terminal Axostyle appears as fine needle like filament .

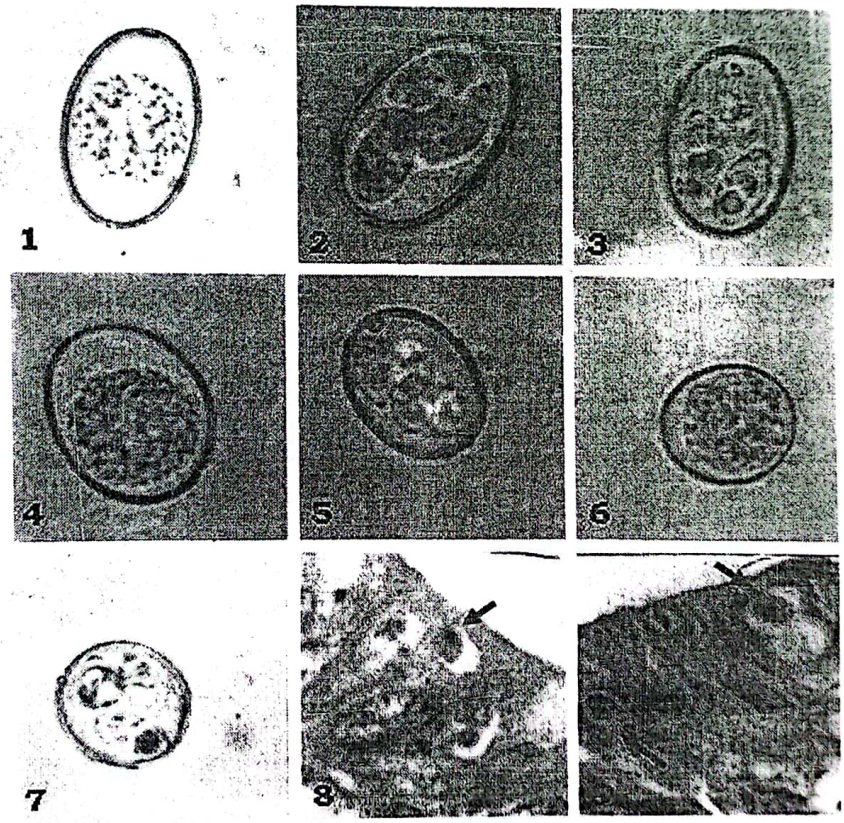

Fig .(1-3):*Eimeria adenoeides* oocyst. (X1000)
Fig. (4-5):*Eimeria meleagrimitis* oocyst. (X1000)
Fig.(6-7): *Eimeria innocua* oocyst . (X1000)
Fig. (8) :*Eimeria adenoeides* trophozoite (X1000)
Fig. (9):*Eimeria adenoeides* schizont (X400)

Fig. (10) :*Cryptosporidium meleagridis* oocyst . (X1000)
Fig. (11): *Cryptosporidium meleagridis* endogenous developmental stages in brush border of tracheal epithelium of experimentally infected rats (X1000)
Double arrow = trophozoites Single arrow =schizonts
Fig.(12-13) :*Tetratrichomonas gallinarum* . (X1000)
Fig.(14-15) :*Tritrichomonas eberthi* . (X1000)

DISCUSSION

Protozoan parasites of turkeys may cause moderate to serious disease or act as predisposing factor for viral infections as viral hepatitis (Wages and Ficken,1989). The present study revealed that the incidence of protozoan parasites in the examined turkeys was 22%. This rate was lower than that mentioned by Elmadawy (2001) who reported that incidence of protozoan parasites in turkeys in Qualubyia province was 31%.

The prevalence of *Eimeria* species in this study was 14 % , this was lower than that of Ozer et al. (1990) and El madawy (2001) who mentioned that the infection rates were 62 % and 20 % respectively while Khalifa(1978) and Bassioni et al. (1979) stated that the incidence of infection with *Eimeria spp.* ranged from 8-20 % in turkey farms. This variations may be attributed to variation in the age, breed and hygienic measures of the examined birds. Three species of *Eimeria; E.adenoeides, E.meleagrimitis* and *E. innocua* were detected in this study, the same species were reported by Elmadawy (2001) in Qaulubyia province while Bond(1967) found that *E. adenoeides , E. meleagrimitis ; E. gallipavonis and E. dispersa* were the most prevalent species of Eimeria in turkey but Utaeva (1973) reported *E.meleagridis* as the most prevalent species .On the other hand, *E.adenoeides, E.meleagridis and E.meleagrimitis* were the most prevalent species in different turkey farms in Egypt (Khalifa ,1978 and Bassioni et al.,1979) while,Ozer et al. (1990) recorded *E.adenoeides, E.meleagridis ; E meleagrimitis* and *E. subrotunda* in Anatolia, Turkey. Such variation may be due to geographical distribution and immune system of the host .

The diagnostic tables of *Eimeria* species in turkeys reported by Boch &Supperer (1977) and McDougald & Reid (1997) concluded that the ellipsoidal oocysts of *Eimeria* might be *E.adenoeides , E.gallipavonis* or *E.meleagridis* , the ovoidal ones might be *E.dispersa , E. meleagrimitis* while the spherical or sub-spherical oocysts might be *E.innocua* or *E.subrotunda . Eimeria adenoeides* characterized by ellipsoidal shape , prominent micropyle and polar granules , these features were similar to that described by Levine (1972) who mentioned that prominent micropyle in the ellipsoidal oocysts is a characteristic feature for *E.adenoeides* ; Pellerdy (1974), Bassioni et al.(1979) and Elmadawy(2001). *Eimeria meleagrimitis* oocysts were ovoid in shape with polar granule as described by Pellerday(1974) and Elmadawy(2001),it should be differentiated from *E. dispersa* by presence of polar granule in the first species according to the table given by Boch & Supperer (1977). *Eimeria innocua* and *E.subrotunda* were spherical in shape , have no polar granules and difficult to be differentiated

morphologically from each other, the present oocyst was identified as *E.innocua* due to its shorter sporulation time than that of *E.subrotunda.* (McDougald & Reid, 1997) .

The incidence of *Cryptosporidium meleagridis* oocysts in the examined turkey was 6% . It was lower than that recorded by Woodmansee et al. (1988) and Elmadawy (2001) which was 38 % and 17.8 % respectively. Also, Sreter et al. (2000) isolated oocysts of *C.meleagridis* from turkey in Hungary. The present morphological features of the detected oocyst were similar to that described by Pellerdy (1974). Experimental infection of white rats with oocysts of *C. meleagridis* revealed the presence of oocysts in the faeces and endogenous developmental stages on the brush border of tracheal epithelium, this means that this species of *Cryptosporidium* was infective to mammals including human . This result agreed with Sreter et al. (2000) who mentioned that the oocyts of *C.meleagridis* were successfully passaged in turkey and were transmitted from turkey to mice . Also, Darabus (1997) stated that oocysts of *C. meleagridis* isolated from chicken proved to be infectious for several species of mammals including mice ,rats , rabbits and cattle .On the other hand, Akiyoshi et al. (2003) found that *C.meleagridis* appears to infect a broad range of hosts including both avian (chicken and turkey) and mammalian (human , piglet , calf and mouse) species . So, the ability of *C.meleagridis* isolate to infect mammals has significant epidemiological implications with regard to human public health in terms of transmission and the risk associated with exposure to birds and possibly other mammals .

Tetratrichomonas gallinarum and *Tritrichomonas eberthi* were detected in one of the examined sample (2%). Similar results were obtained by Honigberg(1961) ; Lee (1972) and Levine (1985) who reported *T. gallinarum* and *Tritrichomonas eberthi* from the lower digestive tract of turkey , while Elmadawy (2001) stated that all examined turkeys were free from trichomonads infection. On the other hand, Badawy et al. (1999) and Elmadawy(2001) reported the Tritrichomonas eberthi from migrant and domestic quails respectively .The morphological features of *Tetratrichomonas gallinarum* and *Tritrichomonas eberthi* were similar to the description based on light microscopic examination of McDowell (1953); Levine (1985); Pecka (1991) and BonDurant and Honigberg (1994).

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