PREVALENCE OF COCCIDIOSIS IN CHICKEN IN SOHAGE GOVERNORATE

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ABSTRACT

Eimeria sp. is one of the most important parasites that cause very high economic loss in poultry farms in Egypt. It causes a disease called coccidiosis. This study aimed to detect the prevalence of Eimeria species in chicken and detection of its pathological effect within the intestinal mucosa. The present study investigate the prevalence of Eimeria sp. in chicken through fecal examination and the diagnosis was based on direct fecal sample examination (unstained wet mount technique) and concentration techniques, followed by sporulation of unsporulated oocyst for identification of Eimeria spp. and finally studying the pathological effect of this parasite in the intestinal mucosa of infected chicken. The total prevalence rate of Eimeria spp. was (66%). The incidence rate in Broiler chickens was (70%) and in Balady was (58%). The highest percent of infection was at the age of (15-30) day (54.3% in Broiler and 72.4% for Balady), and the disease was more prevalent in winter than in summer. The species that were detected are E.acovullina (the highest prevalence rate) followed by E.tenella, E.necatrix followed by E.mitis (lowest prevalence rate). These results indicate that the coccidiosis is a serious parasitic disease that effect on the poultry production in Egypt and control measures should be put in consideration to overcome this disease.

Keywords: Eimeria - prevalence- coccidiosis- oocyst.

INTRODUCTION

Chickens represent the biggest poultry sector that reared intensively and represent a good, cheap and healthy protein source. Among all parasites of poultry, Eimeria are the most economically significant parasite (Blake et al., 2020). Eimeria caused a disease that called coccidiosis, that is widely spread avian disease particularly in chickens either commercial or rural that can seriously affect the development of poultry production (Bachaya et al., 2015).

Coccidiosis is endemic in most of the tropical and subtropical regions where ecological and management conditions are suitable for sporulation of coccidian oocyst and development (Blake and Tomley, 2014). The genus Eimeria is a protozoan within the taxonomic family Eimeriidae, class Coccidia, order Eucoccidiorida and the phylum Apicomplexa. It is obligate intracellular parasites that transmitted to new host cells by invasive extracellular stages.
(sporozoites), which are equipped with a certain structure known as apical complex that aid in penetration of the host cell (Pastor-Fernandez et al., 2018).

There are 9 *Eimeria* species are infective to chickens, identified as *E. acervulina*, *E. mivati*, *E. maxima*, *E. necatrix*, *E. brunetti*, *E. praox*, *E. tenella*, and *E. hagani* (Hamid et al., 2018). *E. tenella* is a highly pathogenic with heavy oocysts producer species, so, this species soon causes coccidiosis in chickens only a few weeks old, while *E. necatrix* is equally virulent but a poor oocysts producer; so it produces the disease in older birds.

The prevalence of coccidia infection primarily affected by the age of the chickens (Sharma et al., 2015), the higher prevalence of coccidiosis at the age of 32–46 days might be associated with the presence of another immunosuppressive disease, such as Gumboro. Young birds are more susceptible and more readily display signs of disease, whereas older chickens are relatively resistant as a result of prior infection (Cervantes et al., 2020).

The life stages of *Eimeria* species develops an endogenous intestinal phase within the host during which three or four rounds of asexual reproduction (schizogony) leading to formation of trophozoit, schizonts (I and II), merozoites (I and II), followed by sexual differentiation into micro and macrogamets (gametogony), fertilization and finally, shedding of the (unsporulated oocysts) within the excreta of the chicken (Francia and Striepen, 2014). Under suitable conditions (25–30°C temperature, moisture, and oxygen) and within 48–72 hours, the process of sporulation begins immediately, yielding sporocysts and infective sporozoites and become infective to another host (sporulated oocysts) (Fanatico, 2006).

*Eimeria* spp causing damage to the cells lining the intestines as it lives and multiply in the cells of the intestinal tract (Arabkhazaeli et al., 2013). The severity of an infection depends on; the age of chicken, *Eimeria* species, number of infective stages that ingested, immune status of the flock and environmental management. Infected birds tend to gathered together, have depression and ruffled feathers. Certain species of *Eimeria* cause dysentery, enteritis, diarrhea, which may be bloody. Emaciation, feed conversion is in lower rate, delayed sexual maturity, drooping wings lead to dehydration and weight loss as well as mortalities. (Abbas et al., 2017).

This study aimed to detect the prevalence of *Eimeria* species in chicken and detection of its pathological effect within the intestinal mucosa and submucosa.

**MATERIALS AND METHODS**

The present study was carried out during the period from August 2020 to August 2021 in the department of Parasitology, Faculty of veterinary Medicine Assiut University.

**Study flocks and samples**

The study was conducted on 150 (intestinal gut samples) taken from broiler-chicken that obtained from different chicken farms and diagnostic labs in Sohage Governorate, Egypt. After birds’ necropsy, the intestine of each bird was dissected out; divided into small intestine and large intestine, then evacuated and scrubed separately into respectively labeled plastic cups, sieved, and preserved using potassium dichromate 2.5% (in ratio of 1:3) and neutral buffer formaline 10%. Mucoid specimens were mixed with several drops of 1% of KOH to avoid trapping of protozoan oocysts during sieving (Gracia, 2001).

**Gross examination**

Intestinal wall, mucosa and serosa were examined for thickening, other pathological changes as haemorrhage, congestion, corregations, tissue debris white spots, and ulceration. Also, blood and abnormal content
were examined if present within the intestinal contents grossly (Garcia, 2001).

Parasitological examination:
a. Direct examination: through
1- Unstained wet mount technique (Garcia, 2001): a drop of the scraped intestinal mucosa put on clean slide and mixed with a drop of 0.9% saline, thoroughly mixed till forming uniform smear. For detection of any coccidian oocysts
2- Concentration technique: Positive Eimeria spp. samples were concentrated using saturated salt floatation concentration technique according to (Cringoli et al., 2010).

b) Sporulation of coccidian oocysts according to (Rao et al., 2013)
For perfect identification, the coccidian oocysts were sporulated. In clean glass petri dishes, the positive faecal samples for Eimeria species were mixed with 2.5% potassium dichromate solution at the depth of 3-5 mm. Petri dishes. The covers of the petri-dishes were lined by moist filter paper and left to stand at room temperature. They were daily aerated and examined to follow up the process of sporulation. The contents of these petri-dishes were concentrated by floatation technique, for identification of the morphological characters of sporulated Eimeria oocysts.

c- Histopathological examination acc. to (Bancroft et al., 1996)
Intestinal specimens were taken from infected chickens, fixed in 10% formaline, dehydrated, cleared, and then embedded in paraffin blocks. 5 μm thicknesses were taken from Paraffin sections, stained by haematoxylin and eosin and examined microscopically.

Statistical analysis
The collected data were analyzed by Statistical Package for Social Sciences v.20 for Windows (SPSS). The significance of differences between the groups were calculated using the Chi-square test for trend analysis (p-value of < 0.001 considered significant).

Ethical consideration
The animal studies were conducted in accordance with the international valid guide lines.

RESULTS
Our study revealed that about 99 birds were infected with Eimeria species from 150 parasitologically examined diseased chickens, with a prevalence rate of 66%. The incidence rate in Broiler chickens was 70% (70/100) and in Balady was 58% (29/50) as shown in Table 1. The examined birds had a history of diarrhea, uneven growth, and bad feed conversion. The post mortem examination revealed enteritis at different localities of the intestine.

In relation to effect of age on the prevalence of infection, there is no infection with Eimeria sp. from 0 to 15 day. The highest percent of infection was at the age of (15-30) day (54.3% in Broiler and 72.4% for Balady), followed by (45.7% in Broiler and 17.2% for Balady) at the age of 30-45day while the lowest infection was detected at the age of higher than 45 day (10.34% in Balady and no infection for Broiler) as shown in Table (2).

In relation to seasonal prevelance of Eimeria spp, the prevalece of Eimeria sp was highest in winter. The percent of infection was (88.5% in broiler and 75% for Balady), followed by autumn (82.6% in broiler and 53.8% for Balady)), while in summer was (54.1% in Broiler and 54.5% for Balady), finally, in spring, the percent of infection was (55.5% in Broiler and 50% for Balady) as shown in (Table 3).

In relation to Mixed infection (infection with more than one species of Eimeria) only about (6/29 from Balady and 16/70 from Broiler were infected with more than one
type of *Eimeria*. the percent of infection is 20.7% in Balady and 22.86% in Broiler). while the rest of examined infected chicken were infected with one species of *Eimeria*. (23/29 from balady and 54/70, the percent of infection is 79.3% in Balady and 77.14% in Broiler) as shown in (Table 4).

The species that were detected are *E.acovullina* (Fig.1) the highest prevalence rate followed by *E.tenella* (Fig.2) followed by *E.necatrix*, (Fig.3) followed by *E.mitis* (lowest prevalence rate) (Fig.4). The species of *Eimeria* were identified according to shape, size and sporulation time as shown in figures.

**Histopathological examination:**
In relation to the pathological changes in the intestinal mucosa, the intestinal epithelial cells showing different changes, there was severe necrosis of intestinal villi and presence of necrotic tissue in the intestinal lumen with inflammatory cell reaction at the intestinal mucosa with different stages of *Eimeria sp* (trophozoites, schizont and oocysts) associated with heavy cellular infiltration at the mucosa and sub mucosal tissues. As shown in the figure (5 and 6).

(A-Eimeria acrevulina non sporulated oocyst) (B-Eimeria acrevulina sporulated oocyst)

(Fig.1) Showing *Eimeria acrevulina* X400 (that are usually oblong oval with two layered wall), Average size: 18 x 15 μ. Sporulation time: 17 h.

(C. E.tenella non sporulated oocys) (D. E.tenella sporulated oocyst)

(Fig.2) Showing *E.tenella* X400 (that are usually ovoid with smooth two layered wall), Average size: 22 x 19 μ. Sporulation time: 1 day.
E. *Eimeria necatrix* non sporulated oocyst

(Fig. 3) showing *Eimeria necatrix* X400 (are usually oblong ovoid with smooth wall without micropyle), Average size: 20 x 17 μ. Sporulation time: 19 h.

G. *E. mitis* non sporulated oocyst

(Fig. 4) Showing *E. mitis* X400 (are usually sub-spherical with smooth wall without micropyle), Average size: 17 x 14 μ. Sporulation time: 20h.

Fig. 5: Photo micrograph of intestine from chicken (Balady) showing severe necrosis of intestinal villi and presence of necrotic tissue in the intestinal lumen X100.
Fig. 6: photo micrograph of intestine from chicken (balady) showing different stages of coccidian parasite associated with hemorrhage and inflammatory cell infiltration X400. The arrow directed to the different stages of Eimeria spp.

Table 1: Total prevalence of *Eimeria* spp in both Broiler and Balady.

<table>
<thead>
<tr>
<th>Eimeria spp</th>
<th>Broiler 70 (70/100)</th>
<th>Balady 58 (29/50)</th>
</tr>
</thead>
<tbody>
<tr>
<td>P value</td>
<td>&lt;0.0001***</td>
<td>0.12</td>
</tr>
</tbody>
</table>

No. /T= number of infected /total number of examined birds

Chi-square for trend analysis was used to compare the proportion of infection in balady and broiler.

Significant difference in broiler group (P< 0.0001.)

Table 2: Prevalence of *Eimeria* spp according age amonge balady and broiler.

<table>
<thead>
<tr>
<th>Age</th>
<th>0-15day</th>
<th>15-30day</th>
<th>30-45day</th>
<th>&gt; 45day</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Broiler (No./T)</td>
<td>0/70(0)</td>
<td>38/70 (54.3)</td>
<td>32/70 (45.7)</td>
<td>0/70 (0)</td>
<td>&lt;0.0001***</td>
</tr>
<tr>
<td>Balady (No./T)</td>
<td>0/29(0)</td>
<td>21/29 (72.4)</td>
<td>5/29 (17.2)</td>
<td>3/29(10.34)</td>
<td>&lt;0.0001***</td>
</tr>
<tr>
<td>Total (No./T)</td>
<td>0/99(0)</td>
<td>59/99 (59.6)</td>
<td>37/99(37.4)</td>
<td>3/99 (3.03)</td>
<td></td>
</tr>
</tbody>
</table>

Chi-square for trend analysis was used to compare the proportion of infection in related to the age

Significant difference between different age group P< 0.0001.

Table 3: Sesonal prevalence of *Eimeria* spp.

<table>
<thead>
<tr>
<th>Seasonal Prevalence</th>
<th>Broiler No./T (%)</th>
<th>Balady No./T (%)</th>
<th>Total No./T (broiler+balady) %</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Winter</td>
<td>23/26 (88.5%)</td>
<td>9/12 (75%)</td>
<td>32/38 (84.4%)</td>
<td>0.013*</td>
</tr>
<tr>
<td>Spring</td>
<td>15/27 (55.5%)</td>
<td>7/14 (50%)</td>
<td>22/22 (53.6%)</td>
<td>0.088</td>
</tr>
<tr>
<td>Summer</td>
<td>13/24 (54.1%)</td>
<td>6/11 (54.5%)</td>
<td>19/35 (54.3%)</td>
<td>0.108</td>
</tr>
<tr>
<td>Autumn</td>
<td>19/23 (82.6%)</td>
<td>7/13 (53.8%)</td>
<td>26/36 (72.2%)</td>
<td>0.019*</td>
</tr>
<tr>
<td>Total</td>
<td>70/100</td>
<td>29/50</td>
<td>99/150 (66%)</td>
<td>&lt;0.0001***</td>
</tr>
</tbody>
</table>

Chi-square for trend analysis was used to compare the proportion of infection in related to the season.

Significant difference between total infection in broiler and balady groups in related to season (P< 0.0001).
Table 4: Single and mixed infection among Balady and Boiler.

<table>
<thead>
<tr>
<th></th>
<th>Single Eimeria infection</th>
<th>Mixed Eimeria infection</th>
<th>Total</th>
<th>P value</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>N</td>
<td>%</td>
<td>N</td>
<td>%</td>
</tr>
<tr>
<td>Balady</td>
<td>23</td>
<td>79.31</td>
<td>6</td>
<td>20.7</td>
</tr>
<tr>
<td>Broiler</td>
<td>54</td>
<td>77.14</td>
<td>16</td>
<td>22.86</td>
</tr>
</tbody>
</table>

Chi-square for trend analysis was used to compare single and mixed infection of infected birds.
Significant difference between single and mixed infection in both Broiler and Balady (P<0.0001).

DISCUSSION

Coccidiosis is worldwide disease problem of intensively reared chickens. Researchers, veterinarians and economists are interested in coccidiosis over many years, due to its severe economic losses among the infected birds, particularly poultry industry where coccidiosis is the most problematic than other livestock. In Egypt, poultry production appears to be the most important of farm animal (FAO, 2006).

The present study revealed that about 99 birds were infected with Eimeria species with a prevalence rate 66%. The incidence rate in Broiler chickens was 70% and in Balady was 58% in relation to the total number examined. The examined birds were diseases and had one or more health problem. The infected birds showed many signs (including dullness, uneven growth, decreased body weight, ruffling and mortality). This incidence of Eimeria species infection among the diseased chickens was indicative to endemicity of the coccidiosis among chickens. Also it gives an idea about irresponsivity of the prevalent Eimeria species in field to the used protective anti-coccidials in feed and also indication to bad management measures.

Our results nearly similar to (Gari et al., 2008) who showed that the prevalence rate of Eimeria was (61.25%) and (Olanrewaju and Agbor 2014) (69%) while it more than that recorded by (Oljira et al., 2012) (20.57%) and (Garbi et al., 2015) (19.5%). While our results where low when compared to (Dinka and Tolossa 2012) in Ethiopia (71.7%), (Al-Quraishy et al., 2009) in Saudi Arabia (80%) and (Lawal et al., 2016) in Nigeria (87.4%). In this study balady chickens was 58%, this result unlike (Ahmed et al., 2003) and (Amer et al., 2010) in Egypt, who recorded the rate of infection (43.9%) and (90%) respectively.

In this study Broiler breeds had high coccidian infection rate (70%). This might be connected to higher stocking densities in broiler production in the study area. The high prevalence also might be due to several factors such as high humidity, time of sampling, poor management and the environment such as accumulation of feces that support the development of Eimeria oocysts.

In relation to effect of age on the prevalence of infection, from 0 to 15 day there is no infection with Eimeria sp. This study agree with studies made by (Etuk et al., 2004), (Amare et al., 2012), and (Dakpogan and Salifou 2013) and disagree with (Badran and Lukešová 2006; Sharma et al., 2015) and (Omer et al., 2011). Absence of infection in age from 0 -15 days this return to protection by maternal immunity, GIT of bird in efficient to crush and digest the oocysts, also, young bird unable to take sufficient number of oocysts to produce infection.
The highest percent of infection was at the age of (15-30) day (54.3% in Broiler and 72.4% for Balady), this nearly agree with (Razmi and Kalideri 2000), (Shirzad et al., 2011), (Ojlira et al., 2012) and (Muazu et al., 2008) and disagree with (Sharma et al., 2015). *Eimeria* spp. can caused infection in all ages of poultry. The age of the chickens is considered as a very important factor in the prevalence of coccidiosis infection. (Badran & Lukesouna 2006; Sharma et al., 2015). Higher prevalence of coccidiosis at the age of 30–46 days might be associated with the presence of another immunosuppressive disease, such as Gumboro (Hachimi et al., 2008; Lanckriert et al., 2010; McDougald & Steve 2008).

In the present study seasonal prevalence showed that the highest infection rate *Eimeria* spp was in winter season (88.5% in Broiler and 75% for Balady), followed by autumn (82.6% in Broiler and 53.8% for Balady), while in summer (54.1% in broiler and 54.5% for Balady), finally, in spring (55.5% in Broiler and 50% for Balady), these results were agree with (Shirley, 1992) and (Ashenafi et al., 2004) who explained that the effect of humidity increase the percent of infection in winter. Also, increasing incidence of coccidiosis in winter is due to increasing the stocking density in winter which may reach to 30% (Lunnden and Thebo 2000) and (Badawy et al., 2000).

Our study was detected that there were different species of *Eimeria* foud within the same bird (Mixed infections) this result agree with (Haug et al., 2008) and (Aarthi et al., 2010), who detected that multiple infections with two or more *Eimeria* spp. were observed in some of the positive cases. In this study, single infection was observed more than mixed infection this parallel to study in Romania (Haug et al., 2008).

The histopathological examination showed different changes (sever necrosis of intestinal villi and presence of necrotic tissue in the intestinal lumen, with inflamartory cell reaction at the intestinal mucosa with different stages of *Eimeria* sp (trophozoites, schizont and oocysts) this in agreement with (Zyan et al., 2017).

In Conclusion, This study indicate that the coccidiosis is a serious parasitic disease that affect the poultry production in Egypt and control measures should be put in consideration to overcome this disease.

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**REFERENCES**


مدى انتشار الكوكسيديا في الدجاج بمحافظة سوهاج

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تعتبر الأيميريا من أهم الطفيليات التي تسبب خسائر اقتصادية عالية جدا في مزارع الدواجن في مصر. وسبب مرض الكوكسيديا. هدفت هذه الدراسة إلى الكشف عن انتشار أنواع الأيميريا المختلفة في الدجاج والكشف عن الأيميريا وتاثيرها المرضي داخل الغشاء المخاطي المعوي. وتبين الدراسة الحالية عن انتشار هذا المرض عن طريق فحص البراز واستخدم التشخيص إلى فحص عينة البراز بالطرق المباشرة وكذلك بتقنيات التركيز لزيادة عدد الطفيل، متبوعًا ببلوغ الطور المتحور لتحديد نوع الأيميريا. وأخيراً دراسة التأثير المرضي لهذه الطفيل على الغشاء المخاطي المعوي للدجاج المصاب. وأوضحت الدراسة أن معدل الانتشار الإجمالي للايميريا بلغ (66%) وبلغت نسبة الإصابة في نجوم التسمين (77%) وفي البلدي (58%). وكانت أعلى نسبة إصابة في سن (15-30) يوم، وكان الانتشار أكثر انتشارا في الشتاء وفي الصيف.شيرب هذه النتائج إلى أن الكوكسيديا مرض طفيلي خطير يؤثر على إنتاج الدواجن في مصر ويتطلب مراقبة تدابير المكافحة للتحكم على هذا المرض.