<u>Original Article</u> Epidemiology of *Eimeria* species in selected broiler farms of Khoy suburb, West Azarbaijan Province, Iran

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ABSTRACT

Intestinal coccidiosis, caused by *Eimeria* species, is an economically-important disease of poultry production industry worldwide. This study was designed to investigate the prevalence of different *Eimeria* species in the farmed broilers of Khoy city, West Azarbaijan, North West Iran. A total of 26 broiler farms of different production capacities were arbitrarily selected and examined in 2013. In each of the farms, Litters of two broilers farms were randomly sampled twice a week and examined. The intensity of infection with each of the *Eimeria* species was assessed on the basis of number of oocysts per gram of litter using Clayton-Lane and McMaster methods. *Eimeria* species diversity was determined by using oocyst sporulation technique in 2% potassium dichromate solution. Results indicated that 23.08% (6/26) of the broiler farms were infected with Eimeria oocysts. The maximum litter infection rate (7.5×10³) was observed in fifth week of the rearing period. The litter infection rate was significantly correlated with kinds of water dispenser, feeder, ventilation, and density. The litters were infected with five *Eimeria* species; *E. maxima* (32.67%) in 6 farms (23.07%), *E. mitis* (24%) in 6 farms (23.07%), *E. acervulina* (18%) in 5 farms (19.23%), *E. tenella* (14.67%) in 4 farms (15.38%), and *E. necatrix* (10.67%) in 3 farms (11.58%). Results of this study uncovered high rates of litter infection with various Eimeria species in the studied farms, suggesting the establishment of firm health management strategies in the region.

Keywords: Broiler, Epidemiology, Litter, Eimeria species, Khoy, Iran

INTRODUCTION

The coccidia comprise of a large group of obligatory intracellular parasites (Duszynski *et al* 1999). The coccidian genus *Eimeria* is common intestinal protozoan parasite of poultry which generally regarded as ubiquitous parasite of poultry (Kaufmann 1996). So far, nine *Eimeria* species have been reported, of which, *Eimeria brunetti*, *E. maxima*, *E. necatrix*, and *E. tenella* are pathogenic, while E. acervulina, *E. mitis, E. praecoxand, E. hagani*, and *E. mivati* are considered as non-pathogenic (Thebo *et al* 1999). Intestinal coccidiosis, caused by various species of *Eimeria*, has become an economically important disease of poultry industry throughout the world (Shirley 1988, Jeurissen *et al* 1996). The disease has led to the intensive economic losses in broiler farms by growth reduction, weight loss, increasing the feed conversion ratio (FCR)

and mass mortalities (Braunius 1980). According to recent estimation in USA, the annual loss by Eimeria infection in poultry-production industry is about \$127 million, similar trend is predictable for many other countries worldwide (Chapman 2009). The Eimeria infection occurs in the poultry by ingestion of the infective oocvsts from litter, soil, food, or water. This parasite commonly invades the epithelial cells of the intestine and causes enteritis with high mortality in period of 3-18 weeks of production (Magner 1991, McDougald & Mattiello 1997). The infected poultry serves as the carrier for spreading the disease in an area mediated by the litter infection (McDougald 1991). Furthermore, poor hygienic and management principles can increase the intensity of the infection (Gross 1985, Jordan 1995). Thus, estimation of the rate of the infected litter could be a reliable criterion for determination of the susceptibility of a broiler farm to the Eimeria infection (Rahbari & Adib Hesami 1995). The recent advances in poultry production in Iran have been remarkable. Khoy municipality in northwestern Iran is a good example for such advancement by having 84 farms growing some 1,700,000 broilers. Till now, no clue of the infection with Eimeria has been reported from the broiler farms of suburban of Khoy, Iran. Thus, this study was aimed to update the knowledge of health conditions in the local broiler farms by investigating the possible contamination with Eimeria species in the litter.

MATERIALS AND METHODS

Litter sampling and oocyst collection. Twenty-six broiler farms in the suburban of Khoy city with different production capacities were randomly selected in 2013. During the course of the poultry production, litters of two broilers farms (100g per 10m2) were taken twice a week. The oocysts of *Eimeria* were detected by using the Clayton-Lane method and their intensity was determined as number of oocysts per gram (OPG) of the litter (Hendrix 1998) and counted by the modified McMaster method (Kaya 2004). The data pertaining to the hygienic and management

conditions of the selected broiler farms were also recorded.

Oocyst sporulation and identification. The collected litters were examined for sporulation and their infection with *Eimeria* species. The litters were suspended in tap water, incubated at room temperature overnight, sieved and dissolved in 2% potassium dichromate solution. The mixture was kept in an incubator at 27 °C for a week (Hendrix 1998). Over 100 oocysts were isolated and examined for *Eimeria* species identification on the basis of their morphometric measures (length, width, shape index) and morphology (shape, wall, color, micropyle structure, oocyst residuum, sporocyst residuum, stieda body) using the identification key provided by Soulsby (1986).

Statisticalanalysis. Statistical analysis was undertaken using a non-parametric Chi-square test, a ttest and One-way ANOVA by SPSS 11.5, SPSS Inc., Chicago, IL, USA. Probability of < 0.05 was regarded as significant.

RESULTS

Prevalence and intensity of the infection. Of all examined broiler farms, the litters of six farms (23.08%) were found to be infected with Eimeria oocysts (Table 2). The maximum litter intensity $(7.5 \times 10^3, \text{ farm no. } 26)$ was observed in the fifth week of the production period (P<0.05). The manual longitudinal ventilation system had significant correlation with litter intensity ($\chi 2=6.686$, P=0.0001). When the chain feeders, manual stud and cylinder feeders system, nipple and bell water dispenser systems were installed in the infected farms, there only was significant difference between litter intensity and the feeders systems ($\gamma 2=13.794$, P=0.008) and the water dispenser ($\chi 2=3.486$, P=0.0001) (Table 1). The addition of vinegar, chlorine dioxide and hydroxide as water disinfectants and coccidiostats as feed additive, and also, flour consumption had no significant effects on the intensity (P>0.05). The intensity had also no correlation with the FCR (P>0.05). However, there was significant correlation between the intensity and broiler stocking density ($\chi 2=2.6$, P=0.0001) (Table 2).

Eimeria species diversity in the litters. Five species of the genus *Eimeria* were identified in the examined farms. *Eimeria maxima* (32.67%) was the most prevalent specie observed in all infected farms, followed by E. mitis (24%) in all infected farms (23.07%), E. acervulina (18%) in 5 farms (19.23%), E. tenella (14.67%) in 4 farms (15.38%), and E. necatrix (10.67%) in 3 farms (11.58%) (Figure 1) (Table 2). Cases of cross-infections with 2 (67%), 3 (66%), and 4 (34.50%) *Eimeria* species were also recorded in the infected farms (Table 2).

DISCUSSION

To implement effective control programs against the infection with Eimeria in poultry-production units, it is useful to have an estimation of the infection intensity in their litters (Braunius 1980, Nematollahi et al 2009). Eimeria species diversity and geographical distribution are directly related to the hygienic and management principles applied against their prevalence (Tavasuli & Pashaei 2004). The investigated broiler farms in the present study showed considerable rates of infection with Eimeria; and the rates were comparable with those reported from the city of Mashad (38%) in North East Iran (Razmi & Kalideri 2000), and Golestan province (36%) in North Iran (Ghaemi et al 2010). The infection rates were much higher in Tabriz (55.96%) in northwestern Iran (Nematollahi et al 2009) and Hamadan (75%) in western Iran (Mehrabi & Yakhchali, 2014). The differences in the infection rates can be attributable to the poultry production technology, health and hygienic management, drug application, immune response and genetic background of the farmed broilers (Yadav & Gupta 2001). Chemotherapy is a commonly-used strategy for the control of coccidiosis in the region (Nematollahi et al 2009, Ghaemi et al 2010). The litter infection was different during the poultry production in the examined broiler farms. The lower litter infection was found in the farms with lower stocking densities, higher FCR

and mortality, and the litter infection rate increased in the last week of the production period. This was in accordance with the reports from England, Argentina, and Iran (Long & Rowell 1975, McDougald & Mattiello 1997, Nematollahi et al 2009, Mehrabi & Yakhchali 2014). In contrast, it was not in line with several other studies (Braunius 1980, McDougald & Mattiello 1997, Chapman & Johnson 1992, Razmi & Kalideri 2000, Tavasuli & Pashaei 2004). These controversial results could be due to different research methodologies. sampling time, managing and production strategies, as well as environmental conditions in different locations. Similar to our findings, E. maxima was the predominant pathogenic Eimeria species in the previous studies in Iran and some other parts of the world (Thebo et al 1988, McDougald & Mattiello 1997, Al-Natour & Suleiman 2002, Nematollahi et al 2009, Adib Nishaboori et al 2010). Mehrabi & Yakhchali (2014) reported four Eimeria species of E. maxima, E. tenella, E. acervulina and E. necatrix from the broiler farms of Hamadan, while the three first species have also been identified in the litter of the broiler farms in different regions of Iran (Razmi & Kalideri 2000, Ghaemi et al 2010). The litter infection with oocysts of Eimeria contributes considerably to the health management in broiler farms. Progressive demand for animal protein, hygienic conditions, and the problem of sustained Eimeria infection at low level are among the major concern in broiler production industry. Recently, broiler farmers in Iran use the feed additive coccidiostats and vaccination to prevent the infection. These seem to have been effective, as in this study, the litter infection was found in the farms where coccidiostats had not been fed to the broilers. Furthermore, the intensity increased with elevations in feed consumption, litter moisture, and broilers body weight. According to Stayer et al. (1995) and Mattiello & McDougald (1997), these are the main factors for the increase in the litter infection with Eimeria. The highest prevalence rate of E. maxima and no clinical signs of coccidiosis in the present study elucidated the presence of subclinical coccidiosis in the

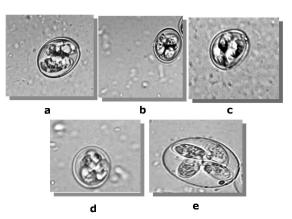


Figure 1. Sporulated oocysts from the litters of the broiler farms in suburban of Khoy, Iran (400×). a, E. tenella; b, E. maxima; c, E. acervulina; d, E. necatrix; e, E. mitis.

Table 1. The hygiene and management characteristics of the broiler farms in suburban of Khoy municipality, Iran

Farm code	Pr.p.	F.p. (×10 ³)	FCR	n/a	W.d.	Fe.	Ve.
21	grower ^a	20	2.4	15 ^{s1}	bell ^{s2}	manual stud ^{s3}	manual longitudinal ^{s4}
22	finisher ^b	10	2.7	14	bell	manual stud	manual longitudinal
23	finisher	3	2.9	15	bell	chain	manual longitudinal
24	finisher	25	2.8	17	nipple	cylinder	manual longitudinal
25	finisher	24	2.9	15	bell	chain	manual longitudinal
26	finisher	30	2.6	16	nipple	cylinder	manual longitudinal

Notes: a, production period from 22 to 35 days ; b, production period from 36 to 47 days; FCR, feed conversion ratio; Fe., feeder system; F.p., farm population; n/a, broiler stocking density, n, number of chicken; a, farm area; Pr.p., production period; Ve., ventilation system; W.d., water dispenser system. s, significance of correlation with intensity: $s_{1,\chi}^{2} = 2.6 (P=0.0001)$ $s_{2,\chi}^{2} = 3.486 (P=0.0001)$ $s_{3,\chi}^{2} = 13.794 (P=0.008)$ $s_{4,\chi}^{2} = 6.686 (P=0.0001)$

Table 2. The prevalence of different Eimeria species in the litters of the broiler farms in suburban of Khoy city, Iran

Eimeria	Prevalence (%)	Si (mean±S		Mixed infection (%)			
species		W	L	SI	2	3	4
Eimeria tenella	14.67	22.8±5.49	18.8±4.81	1.15			
Eimeria maxima	32.67	33.6±8.56	22.4±4.82	1.48		66	34.50
Eimeria mitis	24	16.4±3.3	14.6±2.7	1.03	67		
Eimeria nacatrix	10.67	17.4±3.04	14.8±2.58	1.17			
Eimeria acervulina	18	18.8±1.7	13.4±0.54	1.27			

Notes: L, length; W, width; SI, shape index (L/W).

broiler farms. The litter infection with E. tenella should be taken into consideration as a potential cause of coccidiosis in case of no sanitation and coccidiostats addition, and increased broiler density. As a result, the attention should be paid to improving the hygiene and management principals of the broiler farms. Additionally, further investigations are recommended to evaluate the economic losses due to the subclinical *Eimeria* infection in the region.

Ethics

I hereby declare all ethical standards have been respected in preparation of the submitted article.

Conflict of Interest

The authors declare that they have no conflict of interest.

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