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## RESEARCH ARTICLE

### Detection of *Eimeria* species in Iranian native cattle

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#### Abstract

The aim of this study was to determine the prevalence and diversity of *Eimeria* species in Iranian native cattle from Western Iran. 470 fecal samples were collected randomly in native cattle of Hamedan province. *Eimeria* species was reported in 9.36% of samples. There was a statistical differences among *Eimeria* infection and age groups, unlike to gender and fecal consistency. In conclusion, this study is the first report of prevalence and diversity of *Eimeria* species in Iranian native cattle.

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### Introduction

Bovine coccidiosis is caused by different species of Apicomplexan parasite from the genus *Eimeria* (Almeida et al., 2011). Coccidiosis is responsible for major economic losses in animal husbandry worldwide (Nisar-Khan et al., 2013). Adult animals are usually asymptomatic carriers that often serve as a source of infection for juvenile animals, which are more susceptible to infection (Faber et al., 2002; Abede et al., 2008).

Coccidiosis is commonly a self-limiting disease; and the most signs of bovine coccidiosis is chronic or subclinical (Nalbantoglu et al., 2008). The clinical picture of coccidiosis depends on the innate pathogenicity of different *Eimeria* species. Twelve *Eimeria* spp. have been identified in cattle, worldwide. *E. zuernii* and *E. bovis* are known to be highly pathogenic, causing morbidity and even mortality associated with diarrhea, mucus and blood stains. The other species have been shown experimentally to be mildly or moderately pathogenic, but are not considered important pathogens (Lucas et al., 2006).

*Eimeria* spp. inhibit in the intestinal epithelial cells, leading to mucosa damage and the appearance of clinical signs, malnutrition, weakness, anemia, diarrhea and bloody stool (Yu et al., 2011; Nisar-Khan et al., 2013).

Diagnosis of coccidiosis is by finding oocysts on fecal examination using direct smear, flotation or McMaster's techniques. The number of oocysts per one gram feces (OPG) is helpful in confirming coccidiosis as a cause of clinical disease (Almeida et al., 2011; Nisar-Khan et al., 2013).

The studies on bovine coccidiosis are known in only limited scale in Iran (Yakhchali and Zarei, 2008; Yakhchali and Gholami, 2008; Davoudy et al., 2011). However, there is not published data on coccidiosis in Iranian native cattle.

The main aim of current study was to determine the prevalence and diversity of *Eimeria* species in Iranian native cattle in Hamedan, Western Iran.

### Material and Methods

**Study area and sampling:** Hamedan province by mountainous and mild climate is located in west part of Iran (34.77°N and 48.58°E). It covers an area of 19,546 km<sup>2</sup> and average annual temperature is 11.3°C. This province is economically important for crops and animal husbandry such as cattle in rural regions. In cross-sectional study during 2010 up 2011, 470 fecal samples were collected randomly in native cattle without clinical signs in different

rural regions of Hamedan; kept under the semi-intensive feeding system (Thrusfield, 1997). Fecal samples were directly removed from the rectum of each animal using a disposable examination gloves. The samples were collected in plastic bags, labeled and stored at 4°C until processing. Information about age ( $\leq 1$  and  $> 1$  years old) and gender was obtained from physical examination (Table 1).

**Parasitology examination:** The presence of oocysts in the fecal samples was examined by the flotation method using saturated sodium chloride solution (Yu et al., 2011). The modified McMaster technique was used to quantify the OPG. For sporulation, positive samples were placed in Petri dishes, conditioned with a solution of 2.5% potassium dichromate at room temperature and aired daily for up to two weeks.

The *Eimeria* species were determined based on morphology of oocysts and sporocysts (shape, color, form index, micropyle and its cap, presence or absence of residual, polar granule) and time of sporulation (Soulsby, 1986; Eckert et al., 1995; Yu et al., 2011). There were morphologically characterized at least 10 oocysts of each species for identification (Almeida et al., 2011).

**Statistical analysis:** Statistical analysis was performed by using the software package SPSS version 16.0 for Windows. The differences among variables were evaluated by Chi-square test. A P-value  $\leq 0.05$  was considered statistically significant.

## Result:

Oocysts of *Eimeria* species was found in 9.36% (44/470) of fecal samples (6.73<CI 95%<11.99). The highest rate of *Eimeria* species was determined *E. bovis* (22.7%), followed by *E. zuernii* (18.2%), *E. canadensis* (13.6%), *E. ellipsodalis* (11.4%), *E. alabamensis* (11.3%), *E. pellita* (9.1%), *E. auburnensis* (6.8%), *E. cylindrica* (4.6%) and *E. bukidonensis* (2.3%).

There was a statistical differences among *Eimeria* infection and age groups ( $X^2=32.702$ ,  $P<0.0001$ ,  $DF=1$ , Odds ratio=5.6), unlike to gender ( $X^2=0.866$ ,  $P=0.35$ ,  $DF=1$ ) and fecal consistency ( $X^2=0.325$ ,  $P=0.57$ ,  $DF=1$ ). The overall intensity of infection was calculated between 100 to 1500 OPG (500-1500 in  $\leq 1$  and 100-600 in  $> 1$ yr age). The detailed information is summarized in Table 1.

**Table 1. Prevalence of *Eimeria* species in Iranian native cattle in different age groups, gender and stool consistency in Hamedan, Western Iran.**

	Gender		Age groups (year)		Fecal consistency		Total
	Male	Female	$\leq 1$	$> 1$	Normal	Soft	
<b>No. of sample</b>	225	245	151	319	302	168	470
<b>No. of positive (%)</b>	24 (10.7)	20 (8.2)	31 (20.5)	13 (4.1)	30 (9.9)	14 (8.3)	44 (9.36)

## Discussion

*Eimeria* infection is an important and common disease in cattle; which is responsible for economic losses in worldwide (Davoudi et al., 2011). In previous studies, the occurrence and abundance of *Eimeria* infection in cattle was reported 27-93% in Poland (Pilarczyk et al., 2000; Klovkiewicz et al., 2007), 33% in Hungary (Farkas et al., 2007), 33.3% in Brazil (Almeida et al., 2011), 35-42% in China (Yu et al., 2011), 50% in Pakistan (Nisar-Khan et al., 2013), 68% in Ethiopia (Abede et al., 2008), 20%, 68% and 75% in Turkey (Arslan and Tuzer, 1998; Cicek et al., 2007; Nalbantoglu et al., 2008), 29%, 50% and 52% in South Africa (Matjila and Penzhorn, 2002), and 35% in Tanzania (Chibunda et al., 1997). Also, this rate was reported 21.3% and 18-50% in Kurdistan and East-Azarbaijan provinces in Iran (Yakhchali and Zarei, 2008; Yakhchali and Gholami, 2008; Davoudy et al., 2011).

In our work, the infection rate (9.36%) was lower than other countries; the infection was asymptomatic in all animals similar to studies in Brazil, China and Turkey (Cicek et al., 2007; Almeida et al., 2011; Yu et al., 2011).

Many factors such as the number of ingested oocysts, the presence of a concurrent microbial infection, weather conditions (ambient temperatures and moisture), management in the farms and the functional level of protective immunity may be decisive in whether clinical disease is precipitated or not (Parker and Jones, 1987; Warui et al., 2000).

The prevalence of *Eimeria* spp. in healthy animals implies that they can serve as reservoirs of the infection (Almeida et al., 2011). The native animals are usually resistant to disease and asymptomatic; which is play an important role in transmission of infection to other animals.

During sampling, fecal consistency was normal to soft; there was no significant difference between increase of OPG and fecal consistency (Table 1,  $P>0.05$ ). In other studies from Iran and Brazil, all fecal samples were normal to soft and asymptomatic; despite infection with pathogenic species was observed (Yakhchali and Zarei, 2008; Yakhchali and Gholami, 2008; Almeida et al., 2011).

In our study, the infection rate in  $\leq 1$  yr animals (20.5%,  $P<0.0001$ ,  $OR=5.6$ ) was found higher than  $>1$  yr (4.1%); this is consistent with those of other researchers reporting a strong correlation ( $P<0.05$ ) between the age groups and infection (Chibunda et al., 1997; Abede et al., 2008; Nabantoglu et al., 2008; Nisar-Khan et al., 2013; Almeida et al., 2011; Yu et al., 2011).

Age is a major risk factor in spreading of coccidiosis; morbidity and risk of infection are greater in calves (Abede et al., 2008). Increasing prevalence rate in low age groups may be due to immature immune system and their sensitivity against infection (Matjila and Penzhorn, 2002).

In current finding, infection rate was 10.7% in male and 8.2% in female animals (Table 1,  $P=0.035$ ). By attention to less study of gender and breed, planning and conducting extensive research on the impact and role of different breed and gender in the infection prevalence is highly recommended. Study on interracial breeding and select resistance breeds is very helpful in reducing of economic losses.

In this study, two pathogenic of *Eimeria* spp. including *E. bovis* (22.7%) and *E. zuernii* (18.2%), and seven non pathogenic species including *E. canadensis* (13.6%), *E. ellipsoidalis* (11.4%), *E. alabamensis* (11.3%), *E. pellita* (9.1%), *E. auburnensis* (6.8%), *E. cylindrica* (4.6%) and *E. bukidonensis* (2.3%) were identified in animals. This is in agreement with the finding reported by other researchers (Ernst et al., 1984; Kasiman and Al-Shawa., 1985; Cicek et al., 2007; Farkas et al., 2007; Almeida et al., 2011).

*E. bovis* and *E. zuernii* are known pathogens causing morbidity and mortality in cattle (Friend and Stockdale, 1980; Kennedy and Kralka, 1987; Lucas et al., 2006). A number of authors reported that *E. bovis* was the most prevalent species in cattle, but clinical coccidiosis was not observed in these calves, nor in cows (Ernst et al., 1984; Kennedy and Kralka, 1987; Cornelissen et al., 1995). This finding is in close agreement with the results of our study. The mere present of pathogenic *Eimeria* spp. does not necessarily indicate clinical disease (Warui et al., 2000).

In the present study, OPG in cattle with  $\leq 1$  yr and  $>1$  yr age groups ranged from 500-1,500 and 100-600, respectively. The maximum of OPG levels observed 267,000 in Ethiopia (Abede et al., 2008), 109,000 in Canada (Kennedy and Kralka, 1987), 30,600 in Kenya (Munyua and Ngotho, 1990), and 52,000 in Turkey (Arslan and Tuzer, 1998). Most of animals examined during the current survey had low OPG, suggesting that the infections were usually subclinical. The OPG values over than 5,000 may be pointed out a clinical case (Arslan and Tuzer, 1998).

Different of hygiene conditions and management in farms, breeding of animals, study design and methods, climates and different geographical regions may be the main cause of varied results (Yu et al., 2011).

In conclusion, this study is the first report of prevalence and diversity of *Eimeria* species in Iranian native cattle. The infection with pathogenic *Eimeria* spp. was asymptomatic in all animals; which is responsible to transmission of infection as the carrier and major economic losses in this region. Therefore, integrated strategies should be carried out to prevent and control of *Eimeria* spp. infection in cattle farms.

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