

# Prevalence of thin-walled *Sarcocystis cruzi* and thick-walled *Sarcocystis hirsuta* or *Sarcocystis hominis* from cattle in Iran

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**Abstract** Bovine sarcocystosis is caused by *Sarcocystis cruzi* and is known to cause considerable morbidity and mortality in cattle. This species is distributed worldwide in cattle and is the most prevalent of the *Sarcocystis* species infecting cattle. There is high infection rate of sarcocyst in cattle in Iran, but to our knowledge, there is no study about identification of *Sarcocystis* species. This work aimed to survey prevalence of *S. cruzi* cyst in slaughtered cattle of Isfahan, Iran. In this study, esophageal and diaphragmatic muscles of 100 cattle were collected from Fesaran abattoir of Isfahan and examined for the presence of *Sarcocystis* spp. cysts macroscopically and microscopically. No macroscopic sarcocysts were found in any of the samples. In light microscopy, 89 out of 100 cattle (89%) had thin-walled cysts of *S. cruzi*, while 21 out of them (21%) had thick-walled sarcocysts. In addition to light microscopy, ultrastructural features of the thin-walled cyst confirmed the presence of *S. cruzi*.

**Keywords** *Sarcocystis cruzi* · *Sarcocystis hirsuta* · *Sarcocystis hominis* · Light microscopy · Electron microscopy · Iran

## Introduction

The genus *Sarcocystis* is composed of about 130 species of heteroxenous cyst-forming coccidia with differences in life cycle and pathogenicity. Pathogenic *Sarcocystis* spp. can cause disease in their intermediate host, in particular, in cattle, sheep, pigs, and wild cervids (Guclu et al. 2004).

Cattle are common intermediate hosts of sarcocysts. The prevalence of *Sarcocystis* in adult bovine muscle is close to 100% in most regions of the world (Vangeel et al. 2007). Three species of *Sarcocystis*, *Sarcocystis cruzi*, *Sarcocystis hirsuta*, and *Sarcocystis hominis*, have been recorded from cattle (Saito et al. 1999). Among these species, *S. cruzi* has been identified as the most common sarcocysts around the world. Bovine sarcocystosis is caused by *S. cruzi*, the most pathogenic species, and is known to cause considerable morbidity and mortality in cattle (Wee and Shin 2001).

There is high infection rate of sarcocyst in cattle in Iran; in a recently published study, the prevalence was 100% (Nourollahi Fard et al. 2009), but to our knowledge, there is no study about identification of *Sarcocystis* species.

This work aimed to survey prevalence of *S. cruzi* cyst in slaughtered cattle of Isfahan, Iran.

## Materials and methods

The city of Isfahan is located in the central part of Islamic Republic of Iran. This city is an animal husbandry region and each year exports thousands of cattle to other parts of Iran. From March 2008 to July 2008, esophageal and diaphragmatic muscles of 100 cattle were collected and examined for the presence of sarcocysts macroscopically and microscopically.

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For light microscopy, tissue samples were taken from the esophageal and diaphragmatic muscles and fixed in 10% neutral buffered formalin. They were processed and embedded in paraffin. Sections of 5- $\mu\text{m}$  thickness were cut and stained with hematoxylin and eosin. Some formalin-fixed tissues were postfixed in osmium tetroxide solution, after that the specimens were processed for transmission electron microscopic observation (Saito et al. 1999).

## Results

No macroscopic sarcocysts were found in any of the samples. Histologically, sarcocysts were detected in 92 of 100 cattle. The positive rate was 91% for esophagus and 58% for diaphragm. The infection rate in males and females was 59.46% and 55.55%, respectively. In light microscopy, 89 out of 100 cattle (89%) had thin-walled sarcocysts consistent with *S. cruzi*, while 21 out of them (21%) had thick-walled sarcocysts and could represent either *S. hominis* or *S. hirsuta*. No pathological changes were observed in infected muscle fibers or surround interstitium.

Transmission electron micrographs of thin-walled sarcocysts showed small villar protrusions that were bent approximately 90° to the cyst and oriented nearly parallel to the sarcocyst surface. There was no microtubules in the villar core. The ground substance was found at the base of the protrusions and extended inside the cyst cavity in the form of many septa dividing it into numerous compartments containing the cyst metrocytes and merozoites.

## Discussion

*Sarcocystis* spp. are common parasites with worldwide distribution in man and many species of animals, and infect the skeletal and cardiac muscles (Fayer 2004). The histological distinction between bovine thin-walled sarcocyst (*S. cruzi*) from thick-walled sarcocysts (*S. hirsuta* and *S. hominis*) is simple, whereas the distinction between *S. hirsuta* from *S. hominis* is difficult (Ghisleni et al. 2006).

There are many reports about *Sarcocystis* infections in Iran, and only the presence of sarcocysts in cattle has been reported. To the best of our knowledge, there have been no attempts to identify bovine sarcocysts in Iran. In this study, the prevalence of sarcocysts, *S. cruzi* cyst, and thick-walled sarcocysts was 92%, 89%, and 21%, respectively. In many countries, over 90% of adult cattle have been found infected by *S. cruzi*, and occasionally, this *Sarcocystis* species causes abortion, acute systemic illness, and poor growth (Moré et al. 2008). *S. cruzi* was found in 100% of examined bovine myocardium in Brazil (Santos Da Silva et al. 2002). Experimental and natural studies have shown that

*S. cruzi* is most commonly found in the heart (Gajadhar and Marquardt 1992; Ghisleni et al. 2006). Singh and Sharma investigated prevalence of *Sarcocystis* spp. in cattle in India and differentiated them on the basis of morphological characters and structure of cyst wall. The prevalence of various *Sarcocystis* spp. such as *S. cruzi*, *S. hirsuta*, and *S. hominis* was 41.70%, 9.47%, and 3.79%, respectively (Singh and Sharma 2004).

Several factors including type of animal husbandry system, frequency of final hosts, age of intermediate hosts (Valinezhad et al. 2008), anatomic location, and multiple tissue types for sampling (Woldemeskel and Gumi 2001; Ghisleni et al. 2006) may be associated with prevalence variation of *Sarcocystis* spp. infection. The most important factor to influence this parameter is likely the difficulty to complete the heteroxenous life cycle *Sarcocystis* spp. (Ghisleni et al. 2006). In our study, high infection rate of *S. cruzi* cyst (89%) compared with thick-walled cysts (21%) may be due to the fact that in cattle husbandry fields, there are a lot of sentinel dogs which can contaminate cattle pasture, feed, drinking water, and bedding with *S. cruzi* sporocysts.

No macroscopic sarcocysts were observed in the present study, and this is in agreement with the findings of most previous studies (Okur et al. 1995; Nourollahi Fard et al. 2009). The lack of macroscopic sarcocysts may be due to the fact that such cysts are of feline origin and contact between cattle and cats in this area is not common. The age of the cattle may have been the reason too because it can take years to develop.

Sarcocystosis is not a common disease in humans in Iran. To the best of our knowledge, there is no published study about identification of *Sarcocystis* sporocysts in the feces of human from the test area.

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